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Phan, G.T.

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CHAPTER 7
Pharmacokinetic interactions of antimalarial agents

Phan Trong Giao$^{1,2}$ and Peter J. de Vries$^1$

1 Division of Infectious Diseases, Tropical Medicine and AIDS, Academic Medical Center, Amsterdam, the Netherlands
2 Tropical Diseases Clinical Research Center, Cho Ray Hospital, Ho Chi Minh City, Vietnam

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Abstract

Combination of antimalarial agents has been introduced as a response to widespread drug resistance. The higher number of mutations required to express complete resistance against combinations may retard the further development of resistance. Combination of drugs, especially with the artemisinin drugs, may also offer complete and rapid eradication of the parasite load in symptomatic patients and thus reduce the chance of survival of resistant strains.

The advantages of combination therapy should be balanced against the increased chance of drug interactions. During the last decade, much of the pharmacokinetics and metabolic pathways of antimalarial drugs have been elucidated, including the role of the cytochrome P450 (CYP) enzyme complex. Change in protein binding is not a significant cause of interactions between antimalarial agents. CYP3A4 and CYP2C19 are frequently involved in the metabolism of antimalarial agents. Quinidine is a potent inhibitor of CYP2D6, but it appears that this enzyme does not mediate the metabolism of any other antimalarial agent. The new combinations proguanil-atovaquone and chlorproguanil-dapsone do not show significant interactions.

CYP2B6 and CYP3A4 are involved in the metabolism of artemisinin and derivatives, but further studies may reveal involvement of more enzymes. Artemisinin may induce CYP2C19. Several artemisinin drugs suffer from auto-induction of the first-pass effect, resulting in a decline of bioavailability after repeated doses. The mechanism of this effect is not yet clear, but induction by other agents cannot be excluded. The combination of artemisinin drugs with mefloquine and the fixed combination artemether-lumefantrine have been studied widely, and no significant drug interactions have been found. The artemisinin drugs will be used at an increasing rate, particularly in combination with other agents. Although clinical studies have so far not shown any significant interactions, drug interactions should be given appropriate attention when other combinations are used.
Early diagnosis and prompt treatment of patients with malaria is seen as an important strategy in reducing mortality because it prevents infected individuals progressing to the severe, and possibly fatal, stages of disease. One of the pillars of this strategy is to provide effective antimalarial agents to the primary healthcare level in endemic regions. Because of increasing drug resistance and other factors, the choice of affordable and effective agents has become more and more difficult. Recently, the problem of failing drug therapy was addressed again by reconfirming its high priority on the research agenda

Resistance to almost all antimalarial agents with the exception of the artemisinin drugs has now been reported. There is an urgent need for new antimalarial agents, but new classes of antimalarial drugs are not expected in the near future. While waiting for the discovery of new drugs with novel mechanisms of antimalarial activity, well established and available measures should be taken to safeguard the few compounds available. One of these measures is the appropriate combination of drugs. There are several arguments as to why combination therapy is preferred to monotherapy. However, there are also potential problems regarding drug interactions.

Pharmacokinetic interactions are alterations in absorption, distribution or elimination caused by the combination of drugs. The drug that causes the effect is sometimes referred to as the precipitator or effector and the other as the object or substrate. Interactions between drugs may become significant when the toxic:therapeutic ratio is low and plasma concentrations of the parent compound or of its metabolites may exceed potentially toxic ranges, or when concentrations remain below effective (inhibitory) antiparasitic concentrations.

Precipitators may affect bioavailability by changing absorption or first-pass metabolism. Most drug interactions involving absorption are due to physicochemical incompatibility of 2 drugs (pharmaceutical interactions), but also the composition of the gastrointestinal contents (halofantrine, lumefantrine), motility and intra-luminal enzymatic breakdown (artemisinin drugs) may play a role. Competition for protein binding may lead to displacement of bound drug, but this is rarely of clinical importance. Displacement of protein binding will only be significant when the object drug is 90% or more bound to protein and when the apparent volume of distribution (Vd/F) is very large. In addition, an increase of the unbound drug fraction can increase toxicity in the short term, but this will eventually be balanced by an increased clearance of unbound drug.
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Another mechanism of interaction affects drug elimination. Elimination can be affected by changing uptake into hepatocytes, enzymatic breakdown or of renal excretion. Metabolism reactions are usually divided into type 1 and 2 reactions. Type 2 reactions, such as acetylation and glucuronidation, conjugate compounds making them ready for biliary or renal excretion. Type 1 reactions usually modify the molecular structure of the drug by oxidative reactions. Most type 1 reactions are mediated by the cytochrome P450 (CYP) enzymes, a large family of proteins with a central haem group, classified into families and subfamilies designated by numerals and letters, respectively. The specific isoforms are designated with numerals.

CYP-mediated metabolism mainly takes place in hepatic microsomes, but to small extent also in the intestinal mucosa. Over 30 human CYP enzymes have been identified, of which only the members of families 1, 2 and 3 are involved in the breakdown of drugs, especially CYP1A2, 2C8, 2C9, 2C19, 2D6, 3A4 and 2E1. Induction of enzymatic activity mainly affects CYP1A2, 2C19, 2E1 and 3A4, whereas inhibition mainly affects CYP2D6, 2E1 and 3A4.

Separate genes encode different CYP enzymes. When at least 2 phenotypic enzyme variants can be identified of which the rarest has a prevalence in the population of at least 1%, this is called genetic polymorphism. Genetic polymorphism is known for CYP2D6 and CYP2C19. CYP enzymes can be induced or inhibited by drugs and this is the basis for many drug interactions. Inhibition of a CYP enzyme may transform a rapid metaboliser into a poor metaboliser whereas induction may have opposite effects. For example, erythromycin and ketoconazole are potent inhibitors of CYP3A4, rifampicin (rifampin) induces CYP3A4 and quinidine is a potent CYP2D6 inhibitor. Some nutrients may also affect CYP enzymes, for example grapefruit juice inhibits intestinal CYP3A4. Most of the work on CYP enzymes is based on in vitro enzyme kinetic studies, using cell microsomes, or animal studies. The translation of these findings to the in vivo human situation is often difficult.

Inhibition of elimination can lead to accumulation to toxic drug concentrations, or, in the case of pro-drugs, to ineffective concentrations. Induction of elimination can lead to ineffective concentrations or accumulation of toxic metabolites. Because antimalarial agents are usually administered as short course or single dose regimens, the interactions affecting bioavailability are probably more important than those affecting elimination. Displacement of protein-bound drug may be important during short treatment courses but, as explained, only for drugs with high protein binding.

In this review of the literature on the pharmacokinetic implications of combination therapy for malaria, we will focus on the special properties of the respective agents that could turn
them into potential precipitators or objects of drug interactions. Where available, *in vivo* data from treatment studies in humans will be used. In these studies, different analytical techniques were used and this sometimes has its effect on the interpretation of results. When results are not unequivocal this will be mentioned. Several experimental models, among which are animal models, liver tissue and cellular microsome studies, are also discussed. In most instances the model from which the results originate will be mentioned. Animal (tissue) models do not always reflect the human situation and their results must be interpreted with more caution than human (tissue) studies. However, since there have been few clinical trials performed focusing on pharmacokinetic drug interactions, these experimental findings are often the only source of information.

1. Characteristics of the Main Antimalarial Drugs

There are a few main groups of chemically related antimalarial agents:

- the amino alcohols, including cinchona alkaloids with quinine as the classical example, and synthetic aminoquinolines
- artemisinin with its derivatives
- a group of agents including the antifolates, antibiotics and others.

These agents, which are currently available for treatment or prophylaxis of malaria, will be discussed with the emphasis on their potential interactions with other drugs.

1.1 Cinchona Alkaloids

The bark of the Cinchona tree has long been known for its antimalarial activity. Four active cinchona alkaloids, the laevorotatory quinine, cinchonodine, dextrorotatory quinidine and cinchonine (chemically classified as arylaminoalcohols) can be extracted from the bark. Quinine and quinidine are used in the treatment of patients with malaria, usually as monotherapy but often, because of adverse effects, with a second agent added to shorten the duration of therapy.

The increasing occurrence of drug resistance is another reason why quinine is combined with other drugs nowadays. Cinchonine and cinchonodine are only used in small amounts in the cinchona alkaloid preparation called Quinimax\(^8\). Quinine and analogues exert their antimalarial effect by arresting the multiplication of the mid to late stages of the parasite trophozoites\(^5\). After administration to a patient with malaria there is usually a lag-time before the parasite density starts to decline\(^6\). During that period the parasite count may even
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increase.

1.1.1 Quinine

The pharmacokinetics of quinine have been previously been reviewed in this journal [7]. After oral intake, quinine is well absorbed, irrespective of the formulated base salt, and bioavailability is approximately 80% [8-10]. Bioavailability is very reproducible in healthy individuals, but in malaria variability is larger. The time (tmax) at which the maximum plasma concentration (Cmax) is reached after a single dose ranges from 1 to 4 hours [8,11,12]. The bioavailability of quinine seems to be rather insensitive to external factors but drug interactions certainly have to be studied.

Vd/F is 2.5 to 3 L/kg. Quinine is approximately 70 to 90% bound to plasma proteins, mainly to albumin and the acute phase protein α1-acid glyco-protein [13,14]. Since other antimalarial agents also bind to these proteins, there is a potential competition for binding sites. In malaria, the α1-acid glycoprotein concentration increases, and plasma protein binding of quinine increases to over 90%.

Although this is associated with higher total blood concentrations, a contraction of volume of distribution (Vd) and reduced total drug plasma clearance, the free drug concentrations are less affected [10]. The increased protein binding buffers changes in clearance, which for example may occur in renal failure. It is likely that in malaria this also neutralizes competition for protein binding. However, similar pharmacokinetics have been observed in malnourished children, probably independent of protein binding [15,16]. Changes of protein binding will probably not increase the plasma concentrations of unbound drug to potentially dangerous levels.

The elimination of quinine is mainly by hepatic metabolism. Quinine is a low clearance drug, which undergoes extensive metabolism in the liver, mainly mediated by CYP3A4 and to a minor extent by CYP2C19 [17]. This accounts for approximately 80% of its systemic clearance (CL). Grapefruit juice, a potent inhibitor of intestinal CYP3A4, does not affect the kinetics of quinine, showing that CYP3A4-mediated degradation is located in the liver, not in the gastrointestinal tract [18]. The main metabolite is the biologically active 3-hydroxyquinine but at least 6 other metabolites can be detected [19]. Although metabolism of quinine is mediated by CYP3A4, rifampicin, a potent CYP3A4 inducer, and smoking, which induces CYP1A1 and CYP1A2, both increase elimination of quinine without increasing the formation of the toxic metabolite 3-hydroxyquinine; inversely, it is not likely that inhibition of hepatic CYP3A4 will affect the formation of 3-hydroxyquinine. Apparently several metabolic routes are involved [19].
Elimination of quinine is a mono-exponential process with a mean terminal elimination half-life \( t_{1/2\beta} \) of 10 to 12 hours in healthy volunteers (including children); in elderly individuals the \( t_{1/2\beta} \) is prolonged to 16 to 18 hours. Malaria decreases the metabolic clearance of quinine irrespective of the changed plasma protein binding, so that \( t_{1/2\beta} \) is prolonged to 16 to 18 hours in patients with malaria, a prolongation which was also observed in healthy elderly individuals. Although hepatic clearance is the major route of elimination of quinine, the impact of liver disease is not unequivocal. Acute hepatitis seems to reduce the clearance without affecting plasma protein binding, whereas in chronic liver disease total clearance is unaltered with a reduction of plasma protein binding. Although renal clearance contributes only approximately 20% of total systemic clearance, total quinine clearance is decreased in patients with chronic renal failure, but probably other factors such as altered hepatic clearance and plasma protein binding also contribute to these changes. 3-Hydroxyquinine probably contributes to the adverse effects since it is mainly eliminated by renal clearance, and any renal impairment may increase toxicity.

Drug interactions with quinine as a substrate can increase its toxicity by increasing quinine plasma concentrations or by accumulation of the 3-hydroxy metabolite. Combination with CYP34A4 inhibitors is, at least in theory, to be avoided. Quinine is not a potent precipitator of pharmacokinetic drug interactions. In contrast to quinidine, it is not an inhibitor of CYP2D6. However, while also being a substrate for CYP2C19, competitive inhibition of this enzyme cannot be excluded.

1.1.2 Quinidine

Quinidine is a diastereoisomer of quinine. It is more active against malaria than quinine but the cardiac conduction system is also more sensitive to quinidine. Therefore, the toxic : therapeutic ratio is lower than for quinine, and especially this aspect has to be considered when discussing drug interactions. In most countries, quinidine is mainly used as an antiarrhythmic drug, but where quinine is not available, quinidine is used for the treatment of malaria.

The pharmacokinetic properties of quinidine are comparable to those of quinine, with less plasma protein binding and, as a consequence, somewhat higher free drug plasma concentrations and larger Vd. The clearance of unbound drug is similar. Analogous to quinine, Vd and clearance decrease during malaria. Quinidine is mainly eliminated by CYP3A4-mediated metabolism, but in contrast to quinine, absorption and metabolism to 3-hydroxyquinidine are both inhibited by grapefruit juice, a potent inhibitor of intestinal CYP3A4. Quinidine metabolism is not affected by smoking.
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In humans, quinidine is a potent inhibitor of CYP2D6; it is not a substrate for this enzyme\(^{[25,29,30]}\). Thus, drugs that inhibit CYP3A4 and that are metabolised by CYP2D6 should not be combined with quinidine.

1.2 Mefloquine

Mefloquine is a synthetic antimalarial, available as a racemic mixture. It can be classified as an arylaminoalcohol. Similar to the cinchona alkaloids, it has its greatest activity on the mid to late stage trophozoites. Parasites are cleared almost equally rapid as with quinine therapy\(^{[31]}\). There is no parenteral formulation available, so mefloquine pharmacokinetics have been assessed only after oral administration. There is marked variation in pharmacokinetic properties of the drug, not only between healthy volunteers and patients but also between ethnic groups. In addition, bioavailability may depend on which commercial formulation is used\(^{[32,33]}\). This is an important difference because the dose response relationship is a delicate balance in many endemic areas. Because of this difference, it is difficult to compare pharmacokinetic and efficacy studies.

In general, mefloquine is absorbed slowly; Cmax is reached within 8 to 24 hours. Because of the large variation in bioavailability, this is a potential site for interactions. However, in combination with artesunate it appeared that the timing of drug administration after the onset of recovery is an important determinant for bioavailability, probably more important than the combination with artesunate itself\(^{[34]}\). Food intake increases bioavailability and diarrhoea reduces this\(^{[35,36]}\). Plasma protein binding of mefloquine is also a potential site for drug interactions. It is extensively (90%) bound to plasma proteins. The apparent Vd ranges from 13 to 40 L/kg\(^{[37]}\).

Mefloquine is almost completely eliminated by hepatic biotransformation, with a reported mean t1/2β ranging from 14 to 22 days. Mefloquine, a low extraction drug, is transformed by hepatic microsomal enzymes into its carboxy metabolite, probably mediated by CYP3A4\(^{[38]}\).

It is difficult to interpret these variable results, the more so because mefloquine pharmacokinetics are highly stereo selective and most studies did not address this. The area under the concentration-time curve (AUC) of the (−)-enantiomer is approximately 8.5 times higher and Cmax is 2-fold higher than for the (+)-enantiomer\(^{[39,40]}\). Clearance of the (−)-enantiomer is much slower, with a t1/2β 2 to 3 times longer than for its antipode. It is also very likely that the enantiomers behave differently under different conditions such as malaria and combination therapy.

The effects of malaria on the kinetics of mefloquine are not completely clear. Cmax is
higher in malaria, but with a similar AUC and shorter $t_{1/2\beta}$, suggesting that during the acute disease $V_d$ is contracted, but without a change in plasma protein binding $^{[41]}$. In contrast, it was also observed that in patients with severe malaria, $C_{\text{max}}$ was lower and $t_{1/2\beta}$ was longer than in healthy volunteers $^{[37]}$. Since the residence time of mefloquine is much longer than the time to recover from malaria, the implication of these observations is not clear. The large variability in results also makes it difficult to interpret studies of combinations with mefloquine. However, since mefloquine is combined mostly with artemisinin drugs and since most of these have a very short residence time, potential interactions can be controlled by adequate timing of the dose. Mefloquine is also combined with tetracycline and other agents, and in the past it was also marketed in a fixed combination with pyrimethamine-sulfadoxine (Fansimef$^\text{®}$).

Not much is known about mefloquine as a precipitator of interactions. Mefloquine can decrease bile production in the isolated perfused liver model $^{[42,43]}$. In theory this could interfere with absorption of lipophilic compounds such as halofantrine or lumefantrine. The combinations with these and other drugs will be discussed in section 3.3.

### 1.3 Halofantrine

Halofantrine, a phenanthrene methanol, is an effective antimalarial agent with fast parasiticidal activity on the trophozoite stages of *Plasmodium falciparum*. Its use has been limited mainly because of its effects on cardiac conduction with potentially fatal cardiac arrhythmia. The prolongation of the QT interval in the ECG precludes its use in patients with pre-existing QT syndromes. Combination with other drugs that prolong the QT interval must be avoided.

The pharmacokinetics of halofantrine have been reviewed, and since then not many additional studies have been undertaken $^{[44]}$. The absorption of halofantrine is rather variable. It increases when taken with a fatty meal $^{[45]}$. Upon introduction it was recommended to take halofantrine together with fatty nutrients. To achieve satisfactory treatment responses, especially in Thailand, higher dose regimens were required $^{[46]}$. When cardiac adverse effects were observed $^{[47]}$, the higher dose regimens were no longer recommended and it was advised to take halofantrine on an empty stomach. Since then the use of halofantrine has been limited, and is only used after ECG screening for pre-existing cardiac abnormalities. Thus halofantrine appears to have a low toxic:therapeutic ratio and this is a potential site for drug interactions.

The $t_{\text{max}}$ of halofantrine ranges from 3 hours in healthy volunteers to approximately 16 hours in patients with uncomplicated malaria $^{[48]}$. Halofantrine is metabolised to $N$-desbutyl-
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Halofantrine in the liver, mediated by CYP3A4 and CYP3A5\cite{49,50}. CYP2D6 and CYP2C19 are not involved in the N-desbutylation. Accumulation of halofantrine occurs after repeated doses, suggesting a saturable process of transformation to the desbutyl metabolite. Food intake increases the bioavailability of halofantrine but not of desbutyl-halofantrine\cite{49}. The desbutyl metabolite also has intrinsic activity against malaria parasites and after transformation of halofantrine it still continues to exert its antimalarial effect. Because it is also suggested that the cardiac effects are mediated by halofantrine itself and not by its metabolite\cite{51}, a delay in metabolism of halofantrine, for example by the CYP3A4 inhibitor ketoconazole, would not be advantageous\cite{49}.

The elimination rate of halofantrine is highly variable, with reported mean values of the t\textsubscript{1/2β} ranging from 10 to over 100 hours. However, most of the data were derived from oral intake studies. A formulation for intravenous use has been studied in patients with falciparum malaria. In a 2-compartment kinetic model, the mean t\textsubscript{1/2β} was 14.4 hours in patients with malaria, and 7.5 hours during convalescence\cite{52}. Initial Vd was less than 0.5L/kg.

Thus, the variable oral bioavailability, low therapeutic response in areas such as Southeast Asia and the low toxic : therapeutic ratio with respect to the cardiac effects are the most important factors to be considered for drug interactions. Not much is known about halofantrine as a precipitator of drug interactions. Some combinations with drugs have been studied and these will be presented in section 3.

1.4 Lumefantrine (Benflumetol)

Lumefantrine, formerly called benflumetol, is a racemic fluorene. It is chemically related to halofantrine and the arylaminoalcohols. In contrast to halofantrine, lumefantrine has no effects on cardiac conduction.

Accessible data on the pharmacokinetics of lumefantrine are limited. Most of the preclinical research was done in China as part of the drug development process, without being published. These and complementary data have recently been re-viewed\cite{53,54}. Absorption is rather slow with low and variable bioavailability. In a population kinetic model, the reported median t\textsubscript{max} was 10 hours\cite{55}. Lumefantrine is highly lipophilic and the bioavailability can be increased up to 2-fold by concomitant food intake and this is probably also the cause of increasing bioavailability during recovery from malaria\cite{56}. Lumefantrine is almost completely (>99%) bound to plasma proteins, mainly lipoproteins\cite{57}. In vitro, lumefantrine is metabolised to desbutyl-lumefantrine and excreted via bile and faeces. CYP3A4 is involved in the metabolism. The reported t\textsubscript{1/2β} ranges from 33 hours to 6 days\cite{54,55}.
Not much is known about lumefantrine as an effector. In healthy volunteers the combination of artemether and lumefantrine did not affect the hepatic CYP3A4 activity, as measured by the 6β-hydroxycortisol/cortisol ratio in urine\textsuperscript{58}. Some inhibition of CYP2D6 is mentioned\textsuperscript{53,54} but its clinical significance is not clear. Potential theoretical mechanisms of drug interactions are the high lipophilicity and its consequences for absorption and the extensive plasma protein binding.

1.5 4-Aminoquinolines and Related Compounds

1.5.1 Chloroquine

Chloroquine has been the mainstay of malaria treatment for many years and in large areas of the world it still is, despite an almost global spread of chloroquine resistance. Susceptible parasites are cleared very rapidly from the blood, slightly slower than with the artemisinin derivatives\textsuperscript{59}.

The clinical pharmacokinetics of chloroquine have recently been reviewed\textsuperscript{7,60}. Oral chloroquine is well absorbed in healthy volunteers and in patients with malaria. Vd is enormous, ranging from 100 to 1000L/kg\textsuperscript{61}. Chloroquine is 50 to 60% bound to protein. It is extensively metabolised in the liver to the inactive desethyl-chloroquine, and consecutively to bisdesethyl-chloroquine, 7-chloro-4-aminoquinoline and other compounds. CYP-enzymes probably mediate this, but in the human situation the mechanism is not completely clear. Renal clearance is about 50% of total systemic clearance. Elimination of chloroquine follows multi-compartment kinetics, which also explains the large range of $t_{1/2}\beta$ values reported, ranging from 8 to 58 days\textsuperscript{61,62}. However, the $t_{1/2}\beta$ of chloroquine has little clinical significance in the treatment of malaria but it may contribute to the selection of resistant parasite strains. Intramuscular and subcutaneous chloroquine is very rapidly absorbed and may result in transiently toxic concentrations and therefore the parenteral route is no longer recommended. The pharmacokinetic properties of chloroquine are similar in children and in adults and are not greatly affected by disease severity.

There are \textit{in vitro} data suggesting that chloroquine is an inhibitor of CYP2D6, whether as a substrate or as a non-substrate inhibitor\textsuperscript{63}. However, \textit{in vivo} this effect is less pronounced. In other studies, it was suggested that chloroquine is a CYP3A substrate and/or inhibitor but \textit{in vivo} any inhibitory effect on CYP1A2, CYP2C19, CYP2E1 and CYP3A4 was shown to be absent\textsuperscript{63,64}.

Despite the theoretical potential sites for interaction, chloroquine has been combined with many drugs, intentionally or inadvertently, and significant interactions were not yet reported.
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1.5.2 Amodiaquine

Amodiaquine is also a 4-aminoquinoline. It belongs to the so-called Mannich-base antimalarials, which differ in their dialkylaminomethyl substituent at the C-2' and C6' positions. Amodiaquine is more toxic than chloroquine, which was for a long time the main reason for its limited use. Since amodiaquine has been shown to be active against a significant proportion of chloroquine-resistant parasite strains, there has been a renewed interest in this compound and other Mannich base derivatives with antimalarial activity.

The sparsely documented pharmacokinetics of amodiaquine have recently been reviewed. Amodiaquine can be considered as a prodrug because it is extensively metabolised to the active mono-desethyl-amodiaquine and to the secondary metabolite 2-hydroxy-amodiaquine, which are both eliminated by renal excretion. Absorption is rapid, with the mean tmax of amodiaquine ranging from 0.6 to 1.3 hours in healthy volunteers, followed by tmax of desethyl-amodiaquine at 3 to 5.5 hours. Absorption is slightly retarded in patients with malaria but bioavailability is not affected significantly by malaria. Amodiaquine and monodesethylamodiaquine are over 90% bound to plasma proteins, which is thus a potential site for drug interaction.

Elimination of amodiaquine follows first-order kinetics, with a mean $t_{1/2\beta}$ in healthy volunteers ranging from 5.3 to 7.9 hours. The $t_{1/2\beta}$ of desethyl-amodiaquine ranges from 9 to 18 days. Ketoconazole, a potent CYP3A4 inhibitor, decreases formation of desethyl-amodiaquine in rats but not in human liver microsomes. Since the efficacy of amodiaquine also depends on efficient transformation into its monodesethyl metabolite, there is some concern that combination therapy can antagonise the activity of amodiaquine. This should be studied further.

There are no data about amodiaquine as an effector of drug interactions. Clinical data for combinations with amodiaquine have thus far not shown any significant interactions.

1.5.3 Pyronaridine

Pyronaridine is a bis-Mannich base, an analogue of amodiaquine, with an acridine nucleus instead of a quinoline nucleus. There is no intrinsic cross resistance with chloroquine. The drug is used in China, in combination with other drugs.

The pharmacokinetics of pyronaridine have not yet been extensively investigated. In healthy volunteers a single intramuscular dose was absorbed rapidly with a tmax of 0.66 hours and Cmax of 525µg/L, the $t_{1/2\beta}$ is approximately 64 hours. After oral administration of 600 mg, the reported tmax ranged from 5 to 14 hours.
Pyronaridine may be an alternative first-line drug in areas with significant chloroquine resistance [72]. Further pharmacological studies are required to document disposition and potential drug interactions. From what is being reported from China, however, it seems that the drug can be combined with several other antimalarials.

1.6 8-Aminoquinolines

1.6.1 Primaquine

Primaquine is still the drug of choice for treatment of exoerythrocytic stages of *P. vivax* and *P. ovale*. Primaquine is mostly formulated as a diphosphate salt. It is rapidly and almost completely absorbed with tmax ranging from 1 to 4 hours [39,73,74].

Primaquine is metabolised in the liver to the inactive carboxy-primaquine and several other metabolites. The carboxy metabolite is not toxic but other metabolites are more toxic than primaquine. Metabolism is mainly mediated by hepatic CYP enzymes, but *in vitro* data show that also non–CYP-mediated oxidative processes play an important role [75]. The t1/2β ranges from 1 to 16 hours in patients and healthy volunteers [39,73,76]. There is a report of a time-dependent decline of the AUC after repeated doses in healthy volunteers, the AUC of the last dose being significantly lower and clearance being higher than after the first dose, but these findings have not been confirmed and opposite results have also been reported [76]. Malaria seems to reduce the oral clearance of primaquine [74]. Glucose phosphate dehydrogenase (G6PD) deficiency, the principal risk factor for primaquine-induced haemolysis, does not affect the pharmacokinetics of primaquine [39,74].

Because of the pluriform metabolic pathways, it is difficult to predict when primaquine will act as a substrate for drug interactions. However, the haematological adverse effects, especially in G6PD deficiency, are a matter of concern when considering drug interactions. Toxic metabolites contribute to the adverse effects of primaquine and accumulation of these should be avoided. However, until now, primaquine is mainly used in monotherapy to prevent relapses of tertian malaria or as a prophylactic; drug interactions with other antimalarial agents will not be a common problem in this setting.

Primaquine has not been studied extensively as a precipitator of drug interactions. Studies of combinations with other antimalarials are discussed in section 3.

1.6.2 Tafenoquine

Tafenoquine is a new 8-aminoquinoline, previously referred to as WR-238605. It was developed by the Walter Reed Army Institute of Research [77]. Tafenoquine is absorbed slowly with tmax occurring at 12 hours. The t1/2β is approximately 14 days in humans.
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Although bioavailability in humans is not exactly known, it is estimated that the Vd is extremely large. In vitro, the metabolism of tafenoquine includes CYP-mediated reactions typical for degradation of 8-aminoquinoline lines\(^{78}\). It is not yet completely clear whether tafenoquine, or primaquine, will act as a potent precipitator or object of drug interactions. Until now there are no indications for this, but further studies have to be performed.

1.7 Artemisinin Drugs

Of all known antimalarial agents, the artemisinin drugs have the fastest parasite reduction ratio and the broadest parasite stage specificity\(^{[5]}\). Artemisinin is a sesquiterpene lactone peroxide extracted from the herb Artemisia annua L. With dihydroartemisinin as an intermediate, several derivatives can be synthesised, including the water-soluble artesunate (artesunic acid) and the lipid-soluble artemether and artemotil (artether). These compounds have all been evaluated clinically and they appear to be very well tolerated and effective in clinical practice. Artemisinin and its derivatives express a very powerful and fast antimalarial effect, but because of their rapid elimination and a time-dependent decline of plasma concentrations, recrudescence is a common problem. This is usually avoided by combining them with other antimalarial agents, but extending the duration of monotherapy is also being applied. Several combinations have been used to treat malaria, and no important interactions were noted, but detailed studies into the pharmacokinetic interactions with artemisinin drugs are very limited.

There are indications that the artemisinin drugs may induce neurotoxic effects in experimental animals during extended administration of supra-therapeutic doses. The lipophilic derivatives artemotil, artemether and artemotil appear to be more toxic than the hydrophilic derivatives\(^{[79,80]}\). However, after therapeutic doses in humans no adverse effects have been reported.

The pharmacokinetic properties of the artemisinin drugs have been reviewed recently, but knowledge of this relatively new class of drugs is still expanding rapidly\(^{[81]}\).

1.7.1 Artemisinin

Artemisinin, taken orally, is rapidly absorbed but bioavailability is low, probably due to incomplete absorption\(^{[82,83]}\). t\(_{\text{max}}\) is approximately 2 to 3 hours. Compared to intramuscular injection of a suspension in oil, the relative bioavailability is 32%\(^{[84]}\). Rectal absorption is poor, with bioavailability compared to oral intake approximately 30%\(^{[85,87]}\). Artemisinin is approximately 85% bound to plasma proteins\(^{[88]}\).
Elimination of artemisinin is a rapid mono-exponential process with an \( t_{1/2} \beta \) of approximately 2 to 3 hours. An interesting phenomenon has been observed in multiple dose studies. After repeated doses the plasma concentrations show a decline, with a 6-to 7-fold reduction of the AUC after 6 days of daily administration \(^{[89,90]}\). It is likely that artemisinin induces its own enzymatic breakdown, resulting in increased first-pass effect, but the exact mechanism has not yet been elucidated. For example, it has not been studied whether production of metabolites is increased. The time-dependent decline of artemisinin also occurs after rectal administration, which suggests that the induction should be located in the liver \(^{[85]}\). This decline has also been observed with artemether and with dihydroartemisinin concentrations after oral administration of artesunate.

The single dose pharmacokinetics of artemisinin are similar for healthy volunteers, for patients with uncomplicated malaria and for patients with chronic liver disease, and food intake does not cause significant changes in pharmacokinetics \(^{[82,83,91]}\).

Artemisinin is transformed into inactive metabolites in the liver, mediated by CYP enzymes. In vitro it has been shown that hepatic metabolism is mainly mediated by CYP2B6 and to a lesser extent by CYP3A4 \(^{[92]}\).

The time-dependent kinetics suggest that artemisinin is an inducer of enzymatic degradation processes. This is supported by the finding that artemisinin seems to increase the activity of CYP2C19, but not CYP3A4. However, the time-dependent decline of the AUC of artemisinin was independent of CYP2C19 phenotype \(^{[93]}\). At least one other enzyme, possibly CYP2B6 but not CYP3A4, mediates the self-induced metabolism \(^{[94]}\).

### 1.7.2 Dihydroartemisinin

Dihydroartemisinin was marketed later than artemisinin and the other derivatives, mainly because the physicochemical properties were comparable to those of the parent compound artemisinin. Further, dihydroartemisinin is not water-soluble and hence not suitable for parenteral formulation. However, since the chemical synthesis from artemisinin is rather cheap and because it has a much higher intrinsic activity against *P. falciparum* than artemisinin, it appeared to be attractive to produce dihydroartemisinin for clinical application. Much of the pharmacokinetic information on dihydroartemisinin is known from studies with artemesunate and artemether. These results suffer more (artemether) or less (artesunate) from the fact that the formation rate of dihydroartemisinin from these drugs is not exactly known. There are a few pharmacokinetic studies with dihydroartemisinin taken orally \(^{[95-97]}\). \( t_{max} \) ranges from approximately 1 to 2 hours in healthy Vietnamese as well as in Chinese patients with malaria. Bioavailability of dihydroartemisinin tablets is 2-to 10-fold greater than of
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Artemisinin tablets, and approximately 3-fold greater for suppositories\textsuperscript{[96,97]}. After therapeutic doses, dihydroartemisinin is approximately 47 to 76\% bound to plasma proteins, mainly albumin. The $t_{1/2}\beta$ of dihydroartemisinin is approximately 1 to 2 hours. In the isolated perfused liver rat model, the clearance of dihydroartemisinin was similar to the perfusion rate, which suggests that dihydroartemisinin is a high extraction drug, subject to extensive hepatic metabolism\textsuperscript{[98]}. However, the metabolic fate is not completely clear. It is probably glucuronidated but in rodent liver microsomes dihydroartemisinin is extensively hydroxylated\textsuperscript{[99]}. In the isolated perfused rat liver model, dihydroartemisinin is glucuronidated, mediated by several isoforms of glucuronyl-transferase\textsuperscript{[98]}. Glucuronidated dihydroartemisinin is mainly eliminated by biliary excretion, and by renal excretion for approximately 20 to 30\%. In rodent studies and in the isolated perfused liver rat model, glucuronidation and oxidation processes, with biliary excretion and hepatic clearance, are impaired by \textit{P. berghei} infection\textsuperscript{[98,99]}. Whether this also translates into altered pharmacokinetics of dihydroartemisinin in patients with malaria is a topic for further study.

With respect to interaction with hepatic enzymes, not much is known about dihydroartemisinin, but it seems to be a rather inactive compound. It was shown that NADPH-dependent production of hydrogen peroxide in rat liver microsomes was not enhanced by dihydroartemisinin, in contrast to artepotol and artemelinic acid\textsuperscript{[101]}.

1.7.3 Artesunate

Artesunate (artesunic acid) is a water-soluble derivative of artemisinin. The transformation of artesunate to dihydroartemisinin after oral as well as after intravenous administration is so fast that artesunate can be considered a prodrug. After oral administration, artesunate concentrations are very low\textsuperscript{[102-104]}. The oral bioavailability is 15\% compared with the intravenous route\textsuperscript{[102]}. The mean absorption time of dihydroartemisinin after oral intake of artesunate is slightly longer than 1 hour. After intravenous administration, the $t_{1/2}\beta$ of artesunate is approximately 2.7 min. The reported $t_{1/2}\beta$ of the metabolite dihydroartemisinin is approximately 40 min after oral as well as after intravenous administration of artesunate.

After repeated oral administration of artesunate, a decline of plasma concentrations of dihydroartemisinin was observed\textsuperscript{[104]}. The artesunate concentrations were very low so that it could not be ascertained whether artesunate concentrations also declined. Since after oral administration the activity of artesunate depends mainly on dihydroartemisinin concentrations, the decline is probably a significant contribution to the high rate of recrudescence.

Artesunate is a prodrug of dihydroartemisinin, and thus the possible pharmacokinetic drug
interactions are expected to be the same as with dihydroartemisinin. However, since the bioavailability of oral artesunate probably depends on dissolution in the gastrointestinal tract and intraluminal conversion to dihydroartemisinin, physicochemical (pharmaceutical) interactions should be taken into consideration and studied further.

1.7.4 Artemether

Artemether and the related artemotil are lipid-soluble derivatives of artemisinin. Artemether is available for oral and intramuscular administration. After oral administration, artemether is rapidly absorbed, reaching Cmax after approximately 2 hours. Artemether undergoes extensive first-pass metabolism to dihydroartemisinin in the liver but probably also in the intestinal lumen. Dihydroartemisinin reaches its Cmax after approximately 2 to 6 hours. After oral administration, the AUC of dihydroartemisinin is approximately 2-to 9-fold higher than the AUC of artemether itself, so that dihydroartemisinin probably contributes the majority of the therapeutic effect. After intramuscular and intrarectal administration, absorption is much slower and the dihydroartemisinin : artemether AUC ratio is much lower. In patients with malaria the bioavailability tends to be higher and elimination somewhat slower than in healthy individuals. Artemether is over 90% bound to plasma proteins, mainly to α1-acid glycoprotein and albumin but also to the various lipoproteins. After oral administration, t1/2β ranges from 2 to 4 hours.

After multiple doses, the Cmax of artemether decline whereas dihydroartemisinin concentrations increase. This decline of plasma concentrations is similar to findings with artemisinin and dihydroartemisinin after oral administration of artesunate, suggesting induction of enzymatic activity. The mechanism of this autoinduction is not yet clear. However, since the antiparasitic activity is given by the total concentration of artemether and dihydroartemisinin, and because they both exceed the minimum parasiticidal plasma concentration by many fold, this autoinduction has probably less clinical significance than with artemisinin or artesunate, despite the fact that the rate of recrudescence after monotherapy is comparably high.

Several enzymes, affecting first-pass metabolism and thus bioavailability and possibly also clearance, probably mediate the metabolism of artemether. Bioavailability of artemether can be increased by coadministration of grapefruit juice, suggesting that artemether is a substrate for intestinal CYP3A4. At higher doses, this effect diminishes, which suggests that this intestinal CYP3A4 mediated breakdown is a saturable process. Grapefruit juice does not affect the concentration-time course of dihydroartemisinin. Apparently the amount of dihydroartemisinin reaching the central blood compartment is not affected but the stoichi-
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Ometry of these reactions is not yet clear. Although intestinal CYP3A4 does play a role in the metabolism of artemether, hepatic CYP3A4 activity is not affected as measured by the 6β-hydroxycortisol/cortisol ratio in urine and it is not involved in the time-dependent decline of artemether plasma concentrations [58,10]. CYP2D6 and CYP2C19 are probably not involved in the metabolism of artemether [111].

From these findings, it can be expected that artemether will be involved in drug interactions. Potential mechanisms of interaction affect its CYP-mediated conversion to dihydroartemisinin and the high rate of protein binding. Because of the wide therapeutic index, it is unlikely that the efficacy of artemether is affected by interactions. However, the potential to induce enzyme activity and thus reduce plasma concentrations of other drugs needs further study. One of the possible solutions to avoid drug interactions, particularly in combination with mefloquine, is to administer other antimalarial agents after a short wash-out interval.

1.7.5 Artemotil (Arteether)

Artemotil, previously named arteether, is chemically very similar to artemether. Recently β-arteether was registered in The Netherlands (Arteceft®), renamed generically artemotil; an anomic mixture of arteether has been studied in India. Artemotil is available for intramuscular injection only. Of all the available artemisinin drugs, artemotil has been studied the most in the laboratory setting, but there is only a limited set of data on treatment of humans. After injection, distribution is slow, Cmax occurring after 3 to 12 hours [112]. Plasma protein binding is approximately 80%, to albumin and with a 10-fold higher affinity to α1-acid glycoprotein [113].

In contrast to artemether, artemotil plasma concentrations decline slowly because of the slow release from the intramuscular depot. The pharmacokinetic difference is probably the result of a different formulation of the compound in oil. Artemotil is de-ethylated to dihydroartemisinin. In experimental models this is mediated by CYP isoenzymes, mainly CYP3A4 and to a much lesser extent CYP2B6 and 3A5 [99,114]. P. berghei infection impairs de-ethylation somewhat, but intrinsic (hepatic) clearance is only 4% lower and in the perfused rat liver model this did not alter the pharmacokinetics of artemotil [115].

Potential interactions with the metabolism of artemotil should be studied further, especially those that may reduce the clearance of artemotil. As stated at the beginning of section 1.7, lipophilicity and long residence time of artemisinin drugs are related to neurotoxic effects in animals. Although toxicity has not been shown in humans treated at therapeutic dosages, drug interactions that reduce the clearance of artemotil could possibly cause adverse effects.
Potentially, artemotil is also an enzyme inducer. In rat liver microsomes, artemotil induces NADPH-dependent hydrogen peroxide production, a CYP-mediated oxidative reaction \[^{101}\]. Dihydroartemisinin did not exhibit such an induction. Since artemotil is also a substrate for the CYP enzymes, this may suggest a mechanism for the autoinduction of the artemisinin derivatives, although this has not yet been confirmed for artemotil. When an oral formulation becomes available, this is an important topic of further study.

### 1.8 Antifolates

The folate synthesis of malaria parasites can be interrupted by inhibiting 2 enzymes, dihydropteroate synthetase (DHPS) and dihydrofolate reductase (DHFR). Several point mutations can cause resistance against these 2 types of inhibitors. Mutations coding for resistance occur in almost endemic regions of the world but the pattern and degree of resistance differ \[^{116,117}\]. The interruption of folate synthesis arrests DNA synthesis in the parasite. This explains why the antiparasitic effect has a rather narrow stage specificity and why during treatment the parasites respond later than with the other agents.

#### 1.8.1 Biguanides

Proguanil

Proguanil and chlorproguanil are biguanide derivatives acting as dihydrofolate reductase (DHFR) inhibitors. Therapeutic use of proguanil is limited by the development of resistance of the asexual erythrocytic stages of *P. falciparum*, but it is still used as a prophylactic, often in combination with chloroquine. However, in a fixed combination with atovaquone it has regained interest as a therapeutic agent.

Proguanil is well absorbed, achieving Cmax within 2 to 5 hours \[^{118-120}\]. Proguanil is considered a prodrug, since it is metabolised in the liver to the DHFR inhibitor cycloguanil, but there are also recent indications that proguanil itself enhances the activity of atovaquone \[^{121}\]. Nevertheless, transformation to cycloguanil is rapid; its Cmax occurs 1 hour after the Cmax of proguanil, and Cmax of the inactive metabolite 4-chlorophenylbiguanide occurs a further 1 hour later. The metabolism of proguanil is mediated by CYP3A4 and CYP2C19 \[^{122-124}\]. The latter is a potential site for drug interaction. There is considerable genetic polymorphism of this CYP enzyme, with up to 20% ‘poor metabolisers’ in Asian and African populations \[^{125-127}\]. Poor metabolisers have very low or undetectable plasma concentrations of cycloguanil during prophylaxis. This polymorphism may be the cause of failure of prophylaxis in poor metabolisers, but due to large variability in data a clear association between CYP2C19 activity and efficaciousness of prophylaxis has not yet been demonstrated.
Pharmacokinetic interactions of antimarial agents

The $t_{1/2}$ of proguanil is 12 to 20 hours in patients with malaria and healthy volunteers, but longer in poor metabolisers. The $t_{1/2}$ of cycloguanil is approximately 12 hours.

Chlorproguanil

Chlorproguanil is a chloro derivative of proguanil. It has more intrinsic antiplasmodic activity than proguanil. It is also metabolised to a greater extent to chlorcycloguanil, which has more intrinsic activity than cycloguanil. $t_{\text{max}}$ is approximately 4 hours. The mean reported $t_{1/2}$ ranges from 17 to over 30 hours. Plasma protein binding is over 60% for chlorproguanil and almost 30% for its active metabolite chlorcycloguanil. Chlorproguanil has recently gained more interest, mainly in combination with dapsone. This cheap combination has several advantages. Pyrimethamine-resistant strains retain sensitivity to other DHFR inhibitors, and because of the much shorter $t_{1/2}$ of chlorproguanil than of pyrimethamine, the selection pressure of chlorproguanil for development of resistance is less. Studies on the combination with dapsone, initiated in Kenya, should lead to large-scale application.

1.8.2 Diaminopyrimidines

The diaminopyrimidines are structurally related to the cycloguanides. The main compounds used for the treatment are pyrimethamine and trimethoprim. Both are mainly applied in fixed combinations with sulfonamides.

Pyrimethamine

Pyrimethamine is a DHFR inhibitor. Absorption after oral administration is slow, with $t_{\text{max}}$ ranging from 2 to 12 hours. Pyrimethamine is approximately 90% bound to plasma proteins. Pyrimethamine is mainly metabolised in the liver; there is some renal excretion of unchanged drug. The $t_{1/2}$ ranges from 50 to 106 hours. Pyrimethamine is extensively used in combination with sulfonamides and dapsone for treatment and prophylaxis of malaria and other parasitic diseases such as toxoplasmosis. In all these combinations, drug interactions are not significant.

Trimethoprim

Trimethoprim is also a DHFR inhibitor. It is mostly applied in a fixed combination with sulfamethoxazole, but other combinations are also used. Absorption after oral administration is almost complete, and $t_{\text{max}}$ ranges from 1 to 4 hours. Plasma protein binding is less than 50%. The $t_{1/2}$ is 8 to 11 hours. Trimethoprim is mainly (up to 60%) eliminated unchanged via the urine. A smaller proportion is metabolised in the liver. Most of this is also excreted via the urine, but there is some biliary excretion. Trimethoprim is extensively used in combination with sulfamethoxazole and other agents. Significant interactions have not been
reported. There is a pharmaceutical incompatibility of the intravenous formulation with other agents, including the sulfonamides, but for the treatment of malaria trimethoprim is only used as its oral formulation.

1.8.3 Dapsone

Dapsone (4',4'-diaminodiphenylsulfone) is the most widely used sulfone, but has limited antimalarial activity. It is grouped within the antifolate antimalarials because of its probable inhibition of DHPS. For malaria it is mainly used as a prophylactic in combination with pyrimethamine (Maloprim®), but as stated in section 1.8.1 the combination with chlorproguanil is also being evaluated as a therapeutic combination. Dapsone is rapidly absorbed, with a mean tmax of 2 to 8 hours \[^{[119]}\]. Absorption kinetics are linear over the dose range 75 to 450mg. Concomitant food intake increases Cmax 5-fold. The reported t\(_{1/2\beta}\) ranges from 23 to 70 hours \[^{[119]}\].

Dapsone is metabolised mainly to monoacetyldapsone, and to dapsone hydroxylamine. The major metabolite, monoacetyl-dapsone, is excreted after desacetylation. The intrinsic antiparasitic activity of this metabolite is comparable to that of the parent compound. The acetylation process is subject to interindividual variation. \(N\)-Hydroxylation, a CYP-mediated phase I step, decreases in activity with growing age. There is an inverse relation between acetylation and hydroxylation rates at steady state, but in human liver microsomes the acetylation process does not seem to affect the hydroxylation process\[^{[137,138]}\]. It is thought to mediate the toxicity of dapsone, mainly haemolytic anaemia and methaemoglobinaemia, by formation of the haematotoxic hydroxylamine metabolite \textit{in vitro}.

\textit{In vitro}, CYP3A, CYP2C9 and CYP2E1 have been found to mediate \(N\)-hydroxylation, but not CYP1A2 and CYP2D6 \[^{[139,140]}\]. \textit{In vivo}, CYP3A4 has been identified as an important mediator of this reaction, but not CYP2D6 and CYP2C19 \[^{[141]}\]. Since dapsone is a substrate for CYP3A4, it is object of drug interactions with inhibitors or inducers of this enzyme. For example, inhibiting the hydroxylation of dapsone with cimetidine can reduce toxicity \[^{[142,143]}\]. There is no information about dapsone as a precipitator of drug interactions.

1.8.4 Sulfonamides

The sulfonamides are DHPS inhibitors, and although not necessarily classified under antimalarial agents, in combination with other agents they are used at a very large scale in the treatment of malaria.

The most important sulfonamides in use as antimalarial agents are sulfadoxine, sulfalene (sulfametopyrazine) and sulfamethoxazole. They are mainly used in fixed combinations, sulfadoxine and sulfalene in combination with pyrimethamine (Fansidar® and Metakelfin®,
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respectively) and sulfamethoxazole in combination with trimethoprim as cotrimoxazole. These sulfonamides are absorbed rapidly and almost completely, \( t_{\text{max}} \) occurring at 2 to 6 hours after administration \([144]\). Most sulfonamides undergo metabolic clearance to some extent, mainly by acetylation but also glucuronidation. In contrast to dapsone, the metabolites are not active against malaria. The CYP enzymes do not play a significant role in metabolic breakdown of most sulfonamides. The main difference between the sulfonamides is their elimination rate. Sulfadoxine and sulfalene are eliminated slowly, whereas sulfamethoxazole has a much shorter half-life.

Sulfadoxine is largely bound to plasma proteins, mainly albumin and, like all sulfonamides, it may compete with other drugs for protein binding sites, which makes the sulfonamides potential precipitators of drug interactions \([136, 144]\). In blood, less than 10% of sulfadoxine is acetylated or glucuronidated. Most of it is excreted via the urine in its acetylated form; a minor part is excreted via the stool. The \( t_{1/2\beta} \) is 7 to 9 days. Intramuscular administration increases AUC.

Sulfalene is also readily absorbed and is approximately 60 to 80% bound to plasma proteins. It is mainly excreted via the urine, 5% of which is acetylated \([136]\). The \( t_{1/2\beta} \) is approximately 9 days.

Sulfamethoxazole is also absorbed rapidly, \( t_{\text{max}} \) being reached within 1 to 4 hours \([136]\). Protein binding is stated as 65%. Sulfamethoxazole is mainly eliminated by renal excretion in its acetylated form, and to a lesser extent unchanged.

CYP-mediated degradation is not a major metabolic pathway of the sulfonamides. However there is evidence that certain sulfonamides, including sulfamethoxazole, may inhibit CYP2C9 activity \([145]\). Since trimethoprim may also exert inhibitory effects on CYP2C9, cotrimoxazole should be used prudently in combination with substrates of CYP2C9.

1.8.5 Atovaquone

Atovaquone is one of a series of synthetic hydroxynaphthoquinones found to have potent activity against the AIDS-associated disease *Pneumocystis carinii* pneumonia, toxoplasmosis and malaria. Its pharmacokinetic properties are characterised by relatively poor bioavailability. The oral bioavailability of atovaquone is approximately 23%, which can be increased 3-to 6-fold when taken together with a fatty meal. It is further characterised by low steady-state plasma concentrations and high plasma protein binding. Excretion is almost exclusively through the faeces. There is no significant hepatic metabolism or renal excretion and the \( t_{1/2\beta} \) is long, ranging from 50 to 70 hours \([146]\). Cmax is an important determinant of therapeutic outcome. In patients with HIV, Cmax and AUC tend to be higher when
atovaquone is administered in multiple daily doses instead of single daily doses. However, such a dose-limited absorption was not observed in a population pharmacokinetic study in several groups of patients with malaria [147]. In the same study it was also found that there is no significant interracial difference with respect to pharmacokinetics [147]. In theory, the variable bioavailability and high plasma protein binding expose potential mechanisms for drug interactions.

2. Rationale for Combining Antimalarial Agents

There are several arguments for the use of combinations of drugs for the treatment of malaria. Synergism between drugs or potentiation of their individual effects are the reasons for use in the treatment of an individual patient. Resistance of parasites is the main reason why drugs are being combined for individual patients and why combinations are being advocated on a population level [148].

The development of resistance as a result of drug pressure depends on several factors. These have been reviewed recently [2]. The mutation frequency and the number of mutations required for expression of resistance are genetic determinants. For example, single point mutations may confer DHFR resistance to parasites. Combination with other drugs is beneficial when it increases the number of genes required to express resistance. For example, this is the case with combinations such as sulfadoxine with pyrimethamine. The survival and selection of resistant parasites also depends on the characteristics of the drugs used, such as the pharmacodynamics. The parasite load in a treated patient should ideally be eliminated fast, in order to reduce the chance for mutations to develop, and radically so that no parasites will survive under drug pressure. For example, the fast eradication of the parasite load in patients with malaria and the short $t_{1/2}\beta$ of the artemisinin drugs cause little selective pressure. However, their elimination is so fast that parasites are not easily eradicated and recrudescences – of still sensitive strains – occur. The benefits of the artemisinin drugs are ideally combined with those of other drugs. In contrast, drugs with a very long $t_{1/2}\beta$ such as chloroquine and mefloquine may cause considerable selection pressure, especially when used as monotherapy in high transmission areas. Combinations may also make use of other drug mechanisms than those affected by resistance genes. In the combination of proguanil and atovaquone, for example, proguanil does not seem to act as the prodrug of cycloguanil, but has synergistic activity of its own.

The strong arguments for combination therapy should not undo the awareness that combining drugs may have unforeseen, possibly untoward effects. The interactions of the
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Antimalarial agents have only been sparsely documented. Most currently used combinations proved their benefit without kinetic drug studies ever having been done. Although newer classes of drugs need to be investigated extensively before marketing, covering all the possible drug interactions is difficult. In addition, global marketing of drugs has become more difficult to control and the unregulated use of drugs will increase. All this requires more pharmacotherapeutic expertise from prescribers. To contribute to this, the pharmacology of known antimalarial combinations, including fixed formulations, will be discussed in section 3.

3. Pharmacokinetic Interactions of Antimalarial Agent

In section 1, the pharmacokinetic characteristics of the currently available antimalarials have been discussed and their potential interactions with other drugs, either as object of interaction or as precipitator, have been described. In this section we will discuss what is actually known about interactions of antimalarial agents. Several combinations that are actually in use will not be covered here. That does not mean that these combinations do not show significant interactions. It means that published pharmacokinetic studies were not found. In those instances, the theoretical background in the previous sections, and table I, may serve as a reference.

Drugs can be combined in fixed formulations when their individual administration regimens are approximately similar and when pharmaceutical or pharmacological interactions do not preclude this. When tolerance to the respective drugs or their kinetics change over the treatment course, this may preclude combination in fixed formulations. Some well known currently available fixed formulations are shown in table II.
Table 1. Antimalarial agents and possible mechanisms for pharmacokinetic interactions.

<table>
<thead>
<tr>
<th>Oral bioavailability (%)</th>
<th>Protein binding (%)</th>
<th>Vd f (L/kg)</th>
<th>Cl (L/h/kg)</th>
<th>t 1/2 (h)</th>
<th>fe (%)</th>
<th>Main metabolite</th>
<th>CYP enzymes mediating biotransformation</th>
<th>CYP enzymes affected</th>
<th>Potential and demonstrated pharmacokinetic interactions with other antimalarial agents</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amodiaquine (prodrug) and Monodesethyl-amodiaquine (active metabolite)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80-90 (Amodiaquine)</td>
<td>&gt;90</td>
<td>17-38</td>
<td>5.5-17.1</td>
<td>2-10</td>
<td>5</td>
<td>Monodesethyl</td>
<td>CYP3A4?</td>
<td></td>
<td>In theory: CYP3A4 inhibitors may decrease formation rate of active metabolite, but not yet studied.</td>
</tr>
<tr>
<td>80-90 (Monodesethyl)</td>
<td>(&gt;-90')</td>
<td>(92-285)</td>
<td>(216-756')</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Artemether</td>
<td>43</td>
<td>10-60</td>
<td>0.18-3.0</td>
<td>2-6</td>
<td>0</td>
<td>Dihydroartemisinin</td>
<td>CYP3A4 (also intestinal) and others; not: CYP2C19/2D6</td>
<td></td>
<td>Can induce CYP enzymes</td>
</tr>
<tr>
<td>Artemisinin</td>
<td>30 (compared to IM)</td>
<td>85-88</td>
<td>19-38</td>
<td>4.8-9.0</td>
<td>2-3</td>
<td>&lt;1</td>
<td>Several inactive metabolites</td>
<td>CYP2B6 (CYP3A4)</td>
<td>Induces CYP2C19 (?)</td>
</tr>
<tr>
<td>Only IM</td>
<td>&gt;98</td>
<td>25-30</td>
<td>No estimate (slow release from IM depot)</td>
<td>No estimate</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Artesunate</td>
<td>15</td>
<td>0</td>
<td>2.7 min</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Atovaquone</td>
<td>23</td>
<td>99.9</td>
<td>0.62</td>
<td>0.01</td>
<td>50-70</td>
<td>0</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Chloroquine</td>
<td>80-90</td>
<td>50-64</td>
<td>100-1000</td>
<td>0.10-0.78</td>
<td>40.55</td>
<td>50</td>
<td>(Mono/bis)desethyl</td>
<td>CYP3A4?</td>
<td></td>
</tr>
<tr>
<td></td>
<td>(1.6-13)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Table I (cont.)

<table>
<thead>
<tr>
<th>Aminosalicylate</th>
<th>Protein</th>
<th>Vd/F</th>
<th>Cl [L/h/kg]</th>
<th>t 1/2 [h]</th>
<th>fc [%]</th>
<th>Main metabolite</th>
<th>CYP enzymes</th>
<th>CYP enzymes</th>
<th>Potential and demonstrated pharmacokinetic interactions with other antimalarial agents</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chloroproguanil (prodrug) and Cyclochlor-proguanil (active metabolite)</td>
<td>&gt;60 (&gt;30')</td>
<td>0.48-1.38</td>
<td>17-30</td>
<td>45</td>
<td>Chloroquine : CYP2C19 (genetic polymorphism)</td>
<td>EM (&gt;5%) and PM</td>
<td></td>
<td>In theory: Artemisinin may enhance formation of active cycloguanil. Demonstrated: none</td>
<td></td>
</tr>
<tr>
<td>Dapsone</td>
<td>&gt;90</td>
<td>50-80</td>
<td>1.0</td>
<td>0.04 (0.6)</td>
<td>20-30</td>
<td>20</td>
<td>Monoacetyl- and hydroxyalazine (toxic)</td>
<td>CYP3A4, CYP2C9</td>
<td>for hydroxylation</td>
</tr>
<tr>
<td>Dihydroartemisinin</td>
<td>No estimate</td>
<td>47-76</td>
<td>4-30</td>
<td>1.8-11.4 (30-190)</td>
<td>1-2</td>
<td>&lt;1</td>
<td>Glucuronyl- and other inactive metabolites</td>
<td>No induction of CYP enzymes?</td>
<td></td>
</tr>
<tr>
<td>Halofantrine</td>
<td>&lt;36, increases with fatty food</td>
<td>0.36 (6-8)</td>
<td>0.36-0.48 (10-100)</td>
<td>0</td>
<td>Desbutyl</td>
<td>CYP3A4/5; not CYP2C19/2D6</td>
<td></td>
<td>Not to be combined with quinine or mefloquine. Avoid Cyp3A4/5 inhibitors. Potential effect of other drugs on bioavailability. Demonstrated: none</td>
<td></td>
</tr>
<tr>
<td>Lumefantrine</td>
<td>&gt;99</td>
<td>33-144</td>
<td>0.06-1.2 (1-20)</td>
<td>30-144</td>
<td>&lt;2</td>
<td>Desbutyl</td>
<td>CYP3A4</td>
<td>Inhibits CYP2D6?</td>
<td>In theory. Effects on absorption or competition for protein binding. Demonstrated: none</td>
</tr>
<tr>
<td></td>
<td>Oral bioavailability (%)</td>
<td>Protein binding (L/kg)</td>
<td>Vd/f</td>
<td>CI (L/h/kg)</td>
<td>t β (h)</td>
<td>fe (%)</td>
<td>Main metabolite</td>
<td>CYP enzymes mediating biotransformation</td>
<td>CYP enzymes affected</td>
</tr>
<tr>
<td>----------------</td>
<td>--------------------------</td>
<td>------------------------</td>
<td>------</td>
<td>-------------</td>
<td>---------</td>
<td>--------</td>
<td>-----------------</td>
<td>------------------------------------------</td>
<td>---------------------</td>
</tr>
<tr>
<td>Mefloquine</td>
<td></td>
<td>98</td>
<td>13-40</td>
<td>0.02-0.07</td>
<td>14-22 days</td>
<td>1-9</td>
<td>Carboxy</td>
<td>CYP3A4</td>
<td>Reduces bile production? Not CYP3A4</td>
</tr>
<tr>
<td></td>
<td></td>
<td>98</td>
<td>(0.38-1.2)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Primaquine</td>
<td>75-100</td>
<td>45-65</td>
<td>3-5</td>
<td>0.48-18</td>
<td>1-16</td>
<td>&lt;4</td>
<td>Carboxy and others</td>
<td>CYP3A4 and other CYP enzymes</td>
<td>No significant interactions demonstrated.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(8-30)</td>
<td>(8-30)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Proguanil (prodrug) and Cycloguanil (active metabolite)</td>
<td></td>
<td>75</td>
<td>18-42</td>
<td>1.02-1.38</td>
<td>12-20</td>
<td>40-60</td>
<td>Cycloguanil; EM (17-23)</td>
<td>CYP2C19 (genetic polymorphism), CYP3A4</td>
<td>No significant interactions demonstrated.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(17-23)</td>
<td>(12')</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pyrimefathine</td>
<td>100</td>
<td>87</td>
<td>2.3</td>
<td>0.03 (0.41)</td>
<td>50-106</td>
<td>65</td>
<td>Mainly unchanged</td>
<td></td>
<td>No interactions demonstrated</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(2.5-3.5)</td>
<td>(2.5-5)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Quinidine</td>
<td>71-80</td>
<td>82</td>
<td>2.5-3.5</td>
<td>0.12-0.24</td>
<td>10</td>
<td>15-40</td>
<td>3-hydroxy-</td>
<td>CYP3A4 (also intestinal)</td>
<td>Inhibits CYP2D6</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(2.5-5)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### Table I (cont.)

<table>
<thead>
<tr>
<th>Oral bioavailability (%)</th>
<th>Protein binding (%)</th>
<th>Vd/F (L/kg)</th>
<th>CL (mL/min/kg)</th>
<th>t (h)</th>
<th>fe (%)</th>
<th>Main metabolite</th>
<th>CYP enzymes mediating biotransformation</th>
<th>CYP enzymes affected</th>
<th>Potential and demonstrated pharmacokinetic interactions with other antimalarial agents</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Quinine</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80</td>
<td>80-90</td>
<td>1.8-4.6</td>
<td>0.07-0.24</td>
<td>11-12</td>
<td>10-20</td>
<td>3-hydroxy-</td>
<td>Hepatic CYP3A4</td>
<td>CYP2C19</td>
<td>Protein binding affected by several (disease) conditions. Potential competition with mefloquine for CYP3A4, clinically not significant. Reduces formation of carboxyprimaquine, significance not yet established. Avoid CYP3A4 inhibitors.</td>
</tr>
<tr>
<td><strong>Sulfadoxine</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>70-100</td>
<td>90-95</td>
<td>7-9 days</td>
<td>Almost completely</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Sulfatene</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>70-100</td>
<td>60-80</td>
<td>9 days</td>
<td>Almost completely</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Sulfamethoxazole</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Inhibits CYP2C9 ? Cotrimoxazole potentially inhibits CYP2C9</td>
</tr>
<tr>
<td>70-100</td>
<td>65</td>
<td>0.21</td>
<td>0.02</td>
<td>6-12</td>
<td>14-25</td>
<td>(60% acetylated)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Tafenoquine</strong></td>
<td>No estimate</td>
<td>2550 L</td>
<td>6 L/h</td>
<td>14 days</td>
<td>&lt;1</td>
<td>Carboxy- and others</td>
<td>CYP3A4 and other CYP enzymes</td>
<td>In theory: competition for CYP3A4. Needs further study.</td>
<td></td>
</tr>
<tr>
<td><strong>Trimethoprim</strong></td>
<td>100</td>
<td>-50</td>
<td>1.6</td>
<td>8-11</td>
<td>63</td>
<td>Mainly unchanged</td>
<td></td>
<td>Inhibits CYP2C9 ? Pharmaceutical incompatibility of IV formulation with other formulations</td>
<td></td>
</tr>
</tbody>
</table>

**CL** = systemic clearance; **CYP** = cytochrome P450; **EM** = extensive metaboliser; **fe** = fraction excreted in urine; **IM** = intramuscular; **IV** = intravenous; **PM** = poor metaboliser; **SR** = slow release; **t1/2e** = elimination half-life; **Vd/F** = apparent volume of distribution.
### Table II. Some currently used fixed combinations of antimalarial agents

<table>
<thead>
<tr>
<th>Compounds</th>
<th>Brand name</th>
</tr>
</thead>
<tbody>
<tr>
<td>Artemether 20mg and lumefantrine 120mg</td>
<td>Riamet® tablets</td>
</tr>
<tr>
<td>Atovaquone 250mg and proguanil 100mg</td>
<td>Malarone® tablets</td>
</tr>
<tr>
<td>Chloroquine 100mg and proguanil 200mg</td>
<td>Savarine® tablets (for prophylaxis)</td>
</tr>
<tr>
<td>Quinine 95.3%, quinidine 1.6%, cinchonine 1.1% and cinchonodine 1.1%</td>
<td>Quinimax® 1ml for intravenous/intramuscula r injection</td>
</tr>
<tr>
<td>Sulfadoxine 500mg and pyrimethamine 25mg</td>
<td>Fansidar® tablets and others</td>
</tr>
<tr>
<td>Sulfalene 500mg and pyrimethamine 25mg</td>
<td>Metakelfin® tablets and others</td>
</tr>
<tr>
<td>Sulfamethoxazole and trimethoprim (cotrimoxazole)</td>
<td>Tablets of several brand names</td>
</tr>
</tbody>
</table>

#### 3.1 Combinations with Quinine

Quinine is a very effective antimalarial agent, but its adverse effects and the rather long duration of treatment courses have stimulated its use in combination with other drugs in order to reduce the total dose of quinine. In addition, resistance to quinine has emerged in Southeast Asia and therefore it is recommended to use quinine in combination with other agents – often tetracycline.

There have been many clinical studies on the combination of quinine and other drugs for the treatment of malaria but only few have addressed their pharmacokinetics. As explained, plasma protein binding of quinine is high but it is also subject to variation because of its affinity for α1-acid glycoprotein. Plasma protein binding of quinine is not affected by therapeutic plasma concentrations of chloroquine, mefloquine, primaquine, proguanil or pyrimethamine\[^{14}\]. Other interactions can be expected to affect metabolic clearance. As stated, quinine is transformed into 3-hydroxyquinine mediated mainly by CYP3A4, but other metabolic routes are also followed, including CYP2D6. These enzymes may be subject to induction or inhibition. For example coadministration with rifampicin, a potent CYP3A4 inducer, or cigarette smoking, which induces CYP2D6, increase the metabolic clearance of quinine\[^{149}\]. However, amongst the other antimalarials there are no strong inducers or inhibitors of CYP3A4 and therefore significant interactions are not very likely.

The mixture of cinchona alkaloids marketed as Quinimax\[^8\], which contains small amounts
Pharmacokinetic interactions of antimalarial agents

of other cinchona alkaloids, can be regarded as a combination treatment. It has been suggested that this combination increases plasma concentrations of quinine as well as of quinidine, but these findings have to be confirmed \(^{[150]}\). The bioavailability of quinine when given as rectal Quinimax\(^*\) was found to be 40% of an intramuscular dose of this mixture \(^{[151]}\).

3.1.1 Quinine and Sulfadoxine-Pyrimethamine

In a study of children with severe malaria, a single dose of sulfadoxine-pyrimethamine was administered intramuscularly together with quinine dihydrochloride \(^{[152]}\). The coadministration of sulfadoxine-pyrimethamine did not affect the Cmax, tmax and AUC of quinine, and the results were also similar to the findings in a study in which children with severe malaria were treated with quinine alone \(^{[153]}\).

3.1.2 Quinine and Mefloquine

There are no theoretical data to expect significant interactions between quinine and mefloquine. Both are probably metabolised by CYP3A4 and neither are strong inhibitors or inducers. Plasma protein binding of both is high, which may cause some interaction. The combination was studied in Thailand in adults with malaria \(^{[154]}\). Mefloquine 15mg/kg was given orally together with a 1-hour intravenous infusion of quinine dihydrochloride 10 mg/kg. Plasma concentrations were similar to those in a previous study in which the 2 drugs were given alone to similar groups of patients. However, in another study, when quinine was given with doxycycline and mefloquine \(^{[155]}\), it was found that 4 days after treatment the plasma quinine concentrations were higher and that whole blood mefloquine concentrations were lower was expected from the simulation profiles. Because doxycycline was regarded to have no effect on the pharmacokinetics of quinine \(^{[156]}\), it was concluded that a pharmacokinetic interaction between quinine and mefloquine had occurred. However, even if some pharmacokinetic interaction might be present, there is no significant cardiovascular pharmacodynamic interaction \(^{[154]}\).

Inversely, the situation with quinine as the precipitator and mefloquine as the object has also been studied in vitro \(^{[38]}\). Mefloquine is metabolised to carboxy-mefloquine. In vitro this enzymatic degradation of mefloquine to its carboxy metabolite can be inhibited by quinine, but when extrapolating to therapeutic concentrations this would translate to a modest enzyme inhibition of 40%. When tested in vivo, therapeutic concentrations of quinine did not inhibit the formation of carboxy-mefloquine.

3.1.3 Quinine and Antibiotics

Combinations of quinine and antibiotics, especially tetracycline and clindamycin, are being used but the pharmacokinetic interactions have not been tested. On theoretical grounds it is
not expected that such interactions will occur, unless strong inducers or inhibitors of CYP3A4 are being administered. In African patients with acute falciparum malaria, coadministration of doxycycline did not affect the kinetics of quinine\textsuperscript{156}.

3.1.4 Quinine and Primaquine

As expected, quinine has no significant effect on the kinetics of primaquine, but a decrease of the Cmax and AUC of carboxy-primaquine were observed after coadministration with quinine\textsuperscript{73}. Whether this leads to an increase of other, more toxic, metabolites was not studied but is certainly a matter of concern.

3.2 Combinations with Quinidine

Quinidine is mainly eliminated by CYP3A4-mediated metabolism, which raises concern when it is combined with inhibitors of these enzymes. For example, coadministration with itraconazole increases Cmax and AUC and prolongs the t\textsubscript{1/2}\text{β}\textsuperscript{157}. Itraconazole also inhibits the renal clearance of quinidine, probably by inhibition of renal tubular P-glycoprotein-mediated tubular secretion. In contrast to quinine, quinidine clearance is not affected by smoking.

Quinidine is a potent inhibitor of CYP2D6. It is also a substrate for CYP2D6, but whether CYP2D6 inhibitors can affect the kinetics of quinidine \textit{in vivo} is not clear. The CYP2D6 inhibiting effects of quinidine may affect the kinetics of other drugs. Fortunately, CYP2D6-mediated degradation is not very common, and of the commonly used antimalarials none is metabolised by this enzyme.

Combinations of quinidine with artemisinin derivatives have not been studied in clinical settings, but quinidine, as a CYP2D6 inhibitor, has been used to study the metabolic fate of artemether. It was found that quinidine did not affect the kinetics of artemether in healthy volunteers\textsuperscript{111}.

3.3 Combinations with Mefloquine

Mefloquine has a very long t\textsubscript{1/2}\text{β}. This increases the selection pressure and it is probably the main reason why mefloquine resistance developed soon after introduction in Thailand\textsuperscript{158}. It was combined with sulfadoxine-pyrimethamine in a fixed combination, but because of the high degree of resistance against sulfadoxine-pyrimethamine there was no protective effect for mefloquine. The combination of mefloquine with the artemisinin drugs appears to be a good combination, active against multidrugresistant parasites. It is hypothesised that use of the combination will also retard the development of mefloquine resistance where
Pharmacokinetic interactions of antimalarial agents

Monotherapy has not been used, such as in Viet Nam.

Mefloquine is metabolised to carboxy-mefloquine, catalysed by CYP3A4. In vitro, therapeutic concentrations of artesunate, artemether, sulfadoxine and tetracycline did not inhibit the formation of carboxy-mefloquine \(^{38}\). The combination with quinine has been discussed in section 3.1.2. In general, mefloquine seems to be a rather inactive drug with respect to pharmacokinetic drug interactions. The pharmacodynamic interactions regarding adverse effects are more important.

### 3.3.1 Mefloquine and Primaquine

*In vitro*, there is no or only a small inhibitory effect of mefloquine on carboxy-primaquine formation \(^{42,159}\). *In vivo*, coadministration of mefloquine had no effect on the kinetics of primaquine or its main metabolite carboxy-primaquine in healthy Thai male adults \(^{173}\).

Inversely, mefloquine as an object of interaction with primaquine was studied in human liver microsomes and it appeared that metabolism of mefloquine was somewhat retarded by primaquine \(^{38}\). However, *in vivo* the coadministration of primaquine did not affect the kinetics of mefloquine \(^{160-162}\). There are indications that the combination of mefloquine, primaquine and sulfadoxine-pyrimethamine may shorten the \(t_{1/2}\beta\) of mefloquine when compared to mefloquine with sulfadoxine-pyrimethamine alone, but this is of no clinical significance \(^{162}\).

### 3.3.2 Mefloquine and Halofantrine

Halofantrine is a rapidly acting antimalarial agent. Resistance to halofantrine is often associated with mefloquine cross-resistance and therefore there is little reason to combine the 2 agents when resistance to either of the 2 drugs is prevalent. The pharmacodynamic interactions regarding the cardiac and neuropsychiatric adverse effects are more important. For example, the cardiac effects of halofantrine are enhanced when taken together with mefloquine and this is an extra reason why combination of the 2 drugs is not used routinely \(^{47}\). Nevertheless, the pharmacokinetic interactions between mefloquine and halofantrine have been addressed in a single study using the isolated perfused rat model \(^{43}\). Mefloquine pretreatment did not affect the kinetics of halofantrine, except that there was some increase of clearance of the higher doses of halofantrine. Mefloquine diminished bile production and it was suggested that this cholestasis is a potential mechanism of interaction affecting absorption or excretion of halofantrine. To our knowledge this has no *in vivo* human analogue.

### 3.3.3 Mefloquine and Artemisinin

In the single study on the pharmacokinetics of artemisinin combined with mefloquine, it
was observed that mefloquine coadministration to patients with uncomplicated falciparum malaria increased the AUC of artemisinin by one third to a half, contracted Vd and lowered clearance without affecting $t_{1/2}$ \[163\]. Since the metabolic fate of artemisinin is not yet completely known, the explanation for this observation is not yet clear.

### 3.3.4 Mefloquine and Dihydroartemisinin

Mefloquine did not affect the kinetics of dihydroartemisinin in healthy Thai males and patients with malaria \[164,165\]. Inversely, dihydroartemisinin did not affect the kinetics of mefloquine.

### 3.3.5 Mefloquine and Artesunate

There are suggestions \[166,167\] that artesunate and artemether can affect the pharmacokinetics of mefloquine when given together. The theoretical grounds for such an interaction are not known and these findings should be considered as preliminary. *In vitro*, artemether or artesunate do not affect mefloquine kinetics \[38\]. The suggestion that there is an interaction *in vivo* is based on the finding that in patients coadministration with artesunate was associated with a decrease of the mefloquine Cmax, an increase in the metabolic clearance and an expansion of Vd \[166\]. These findings were explained by competition for protein binding sites. In view of the extreme short residence time of artesunate, this is unlikely.

A possible interaction between the 2 drugs was not found in a large study in Thai children with falciparum malaria; it was found that the AUC of mefloquine on day 0 was also lower than the AUC on day 2 when artesunate was given for 3 days \[34\]. The authors suggested that the recovery from malaria was the main cause of the increased bioavailability of mefloquine. Food intake, as suggested in a previous study, was not a significant effect. However, in a study comparing adult Thai patients and healthy volunteers, malaria only retarded absorption somewhat but AUCs were not different \[36\]. Finally, in another study there was no difference between single dose and 2 divided dose regimens \[168\]. These conflicting data do not provide evidence for an interaction between artesunate and mefloquine. In addition, if there were such an interaction between artesunate and mefloquine than a comparable effect would be expected between dihydroartemisinin (because oral artemate is the prodrug of dihydroartemisinin) and mefloquine, but this was not found in a recent study \[164\].

### 3.3.6 Mefloquine and Artemether

*In vitro*, artemether does not affect mefloquine kinetics \[38\]. But, in analogy with the findings with artesunate, in a small study in patients with uncomplicated malaria the mefloquine AUC was slightly lower when given 24 hours after artemether than when given
Pharmacokinetic interactions of antimalarial agents

alone \(^{[167]}\). As for the interaction between mefloquine and artemesunate discussed in section 3.3.5, the suggested interaction between artemether and mefloquine has to be confirmed in studies dedicated to this question. In addition, oral artemether is a prodrug of dihydroartemisinin, there is no interaction between mefloquine and dihydroartemisinin, and possible interactions with mefloquine are probably not clinically important since efficacy is determined by the combined activity of artemether and dihydroartemisinin. However, it seems rational to delay the administration of mefloquine until initial recovery has been ensured by one of the artemisinin drugs.

### 3.3.7 Mefloquine and Co-artemether (Artemether and Lumefantrine)

The combination of artemether and lumefantrine (co-artemether) is a recently introduced fixed drug combination (Riamet\(^*\)). Because it is likely that co-artemether will be administered soon after or even during therapeutic or prophylactic use of mefloquine, this triple combination was studied in a very nice example of a pharmacokinetic interaction study in healthy volunteers \(^{[58]}\). It was shown that mefloquine administration caused a decrease of lumefantrine bioavailability. It was suggested that a mefloquine-mediated reduction of biliary excretion could contribute to this mechanism. However, in view of the large variability of lumefantrine absorption, the interaction is unlikely to be of clinical importance. There was no interaction between co-artemether and mefloquine with respect to elimination. Since there was also no induction of CYP3A4 activity, it can be concluded that although metabolism of the 3 drugs is mediated by CYP3A4 there is no inhibition of metabolism by competition.

### 3.3.8 Mefloquine with Sulfadoxine-Pyrimethamine

This well-tolerated combination has been used, mainly in Thailand, for several years as a fixed formulation (Fansimef\(^*\)) but did not prevent the development of mefloquine resistance, probably sulfadoxine-pyrimethamine resistance was already prevalent. The combination was not used in the regions where sulfadoxine-pyrimethamine was still effective. There is not much interest now in this fixed combination, and the increase of sulfadoxinepyrimethamine resistance will only support this. In theory, displacement of plasma protein binding by sulfadoxine is a potential source for interactions, but the impact of altered protein binding is usually small. It could possibly increase free mefloquine concentrations and increase adverse effects, but this is not supported by clinical experience. A study has addressed the pharmacokinetic interactions in patients with malaria and it was found that coadministration of sulfadoxine-pyrimethamine does not change the kinetics of mefloquine \(^{[162]}\).

### 3.3.9 Mefloquine with Tetracycline

The interaction between mefloquine and tetracycline has been investigated in a single study
The Cmax of mefloquine increased by approximately one-third when administered together with doxycycline. The t1/2β of mefloquine decreased by approximately one-quarter and Vd decreased by approximately one-third when given together with tetracycline. This is possibly explained by reduced bioavailability and reduced enterohepatic recycling. However, the results suffered from a large variability and it is not clear if there is a clinical consequence.

3.4 Combinations with Chloroquine

Chloroquine has been administered to so many patients under such a wide variety of conditions and comedications that clinically significant interactions would have been documented if there were any. In this vast experience, chloroquine has always performed as a well tolerated drug which can be combined with other antimalarial agents. Perhaps this is the reason that little study has been made of pharmacokinetic interactions with chloroquine. For drugs with a very rapid elimination, such as most of the artemisinin drugs, sequential timing of administration can circumvent potential interactions. However, combination with other drugs with long residence times should be studied.

Nevertheless, the fast reduction of the parasite load after chloroquine could be a valuable contribution to combination therapy, but the long t1/2β would still cause substantial pressure favouring chloroquine-resistant strains. Moreover, because of the currently widespread resistance to chloroquine, combination with other agents often has to be regarded as monotherapy and thus has little protective effect for the other drug. When in the future the degree of chloroquine resistance has decreased as the result of discontinuation of usage, then reintroduction should be in the form of combination therapy.

Another approach to overcome resistance is to combine chloroquine with non-antimalarial agents that reverse chloroquine resistance. For most of these agents this would require supratherapeutic, potentially toxic, doses. However, pretreatment with chlorpheniramine, a histamine H1 receptor antagonist, is used in combination with chloroquine, mainly to alleviate chloroquine-induced pruritus in black skin. It was shown that an anti-pruritus dosage of chlorpheniramine significantly increased the bioavailability of chloroquine by 70% in Nigerian children with uncomplicated falciparum malaria. The mechanism is unexplained and the sample size was small. Confirmation of these findings is required. Supratherapeutic doses of chlorpheniramine are required to reverse drug resistance.

The combination of chloroquine and proguanil has been studied mainly because it is used in prophylaxis, which implies long term administration to healthy volunteers. Chloroquine and proguanil have been used in combination as prophylaxis against P. falciparum for many
Pharmacokinetic interactions of antimalarial agents

years. In view of this, it is surprising that not much clinical data on possible interactions have been published. However, from the studies which documented the single capsule formulation (Savarin*), it is known that the pharmacokinetics of both agents are not affected by coadministration, although the combined formulation is absorbed a little more slowly than separate tablets [171].

3.5 Combinations with Primaquine

Primaquine is mainly used to prevent relapses of tertian malaria, and recently it has regained interest as a prophylactic drug. In these conditions it is not likely to be combined with other antimalarial agents. But, since primaquine is also used in combination therapy in some regions, drug interactions should be taken into consideration. The clinically most significant interactions would be those that increase the formation or accumulation of toxic metabolites. In vitro studies have shown that metabolism to carboxy-primaquine is inhibited by ketoconazole, a potent CYP3A4 inhibitor. Whether this also leads to increase in other, toxic, metabolites is not known. In vitro studies with human liver microsomes did not indicate any effect of quinine, artemether, artesunate, halofantrine or chloro-quine [159]. As stated in section 3.3.1, there is some in vitro interaction between mefloquine and primaquine, without clinical consequence.

3.6 Combinations with Artemisinin Derivatives

Since the problem of drug resistance will inevitably increase the use of the artemisinin drugs in the coming decade, it is important to consider interactions with all antimalarials and also other drugs. The great advantage of the artemisinin drugs is their rapid reduction of the parasite load. Because of the short $t_1/2\beta$ of most artemisinin drugs, recrudescences occur after monotherapy. In combination with other drugs, the short and powerful activity may reduce the selection pressure. Despite these advantages, there is only limited experience with their combined use with antimalarials other than mefloquine. The combination with mefloquine was discussed in section 3.3.

3.6.1 Artemether and Lumefantrine (Benflumetol)

This combination was based on the principle of combining a short half-life artemisinin derivative (artemether) and a long half-life antimalarial drug (lumefantrine), in a fixed artemether and lumefantrine ratio of $1 : 6$. This combination, called coartemether, is well tolerated and effective in the treatment of uncomplicated and drug resistant P. falciparum malaria.
The pharmacokinetics of this combination was recently reviewed\textsuperscript{[54]}. The pharmacokinetics of the 2 drugs given in combination is not substantially different from those of the components given individually. The combination is associated with a slight delay of absorption, but the total AUC did not change and the time-dependent decline of artemether concentrations and increase of dihydroartemisinin concentrations was also observed with the combination\textsuperscript{[107]}.

### 3.6.2 Artesunate and Atovaquone-Proguanil

A single study has looked into the combination with atovaquone-proguanil\textsuperscript{[172]}. In theory this seems a rational combination, but further studies have to confirm this. The pharmacokinetics of the combination atovaquone-proguanil were not affected by coadministration with artemunate.

### 3.6.3 Artemether and Pyrimethamine

The combination of artemether and pyrimethamine is not a popular combination and it is not likely to be in the near future. However, the combination has been studied in healthy Thai males. No significant differences of artemether kinetics were observed. The Cmax of pyrimethamine increased by almost half and Vd contracted by approximately 13\textsuperscript{[133]}%. The explanation is not clear; it is possible that there is some interaction with respect to absorption or protein binding.

### 3.6.4 Combinations with Artemotil

The effect of quinine, quinidine, mefloquine, halofantrine and chloroquine on the de-ethylation of artemotil was studied in human liver micro-somes\textsuperscript{[114]}. Halofantrine was the strongest inhibitor of this process, probably by competitive inhibition of CYP3A4, but the quantitative effects were small. Halofantrine would at therapeutic concentrations inhibit 20\% of enzyme activity. Mefloquine and quinidine would inhibit less than 10\%. Thus, in the clinical setting it is not likely that the de-ethylation of artemotil is greatly affected. However, artemotil may competitively inhibit the metabolism of other drugs. This is yet to be studied.

In conclusion, the artemisinin drugs are ideally suited for combination therapy. Regarding metabolic interactions, dihydroartemisinin seems to be the least active compound. Current experience has shown that combinations with artemisinin are effective and well tolerated, but before they are marketed globally, and thus become combined with other available drugs, their possible interactions should be studied extensively. In particular, artemotil, with its sustained plasma concentrations, has to be studied further.
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3.7 Combinations with Antifolates

The rationale for combining the 2 types of folate synthesis inhibitors is their synergistic activity and the fact that different genes express resistance, which reduces the chance of selection of resistant strains. A large difference in $t_\frac{1}{2}$ of the components of the combination, however, may still increase the chance of selection of resistant strains. Resistance to these inhibitors has developed in most endemic areas of the world, but the degree of resistance differs.

3.7.1 Combinations with Biguanides

Proguanil and chlorproguanil have recently regained interest as therapeutic agents in combination with other antimalarial drugs. From a pharmacokinetic point of view, it is not expected that they will interact greatly with other agents. Their activity depends largely on CYP2C19-mediated transformation into the active cycloguanil derivatives, but proguanil itself also has intrinsic antiparasitic activity, especially in synergy with atovaquone. The biguanides can be considered well tolerated drugs in combinations, either as precipitators or as substrates.

The combination of a biguanide and dapsone has recently regained interest as a treatment for malaria. In healthy volunteers, proguanil has no effect on the kinetics of dapsone. It is unknown whether proguanil has any effect on the kinetics of monoacetyl-dapsone, the main metabolite and which is probably responsible for the adverse haematological effects of dapsone.

The combination of chlorproguanil and dapsone is a rational alternative, especially where sulfadoxine-pyrimethamine resistance is prevalent, such as in East Africa. The pharmacokinetics of the combination chlorproguanil-dapsone in Kenyan children with uncomplicated falciparum malaria did not suggest a significant pharmacokinetic interaction.

3.7.2 Combinations with Atovaquone

The combination of proguanil and atovaquone was studied for use against multidrug-resistant falciparum malaria. In vitro, proguanil has a specific synergistic effect with atovaquone, which is not so much dependent on its active metabolite cycloguanil but rather on its specific biguanide effect. Therefore, the combination is effective in cycloguanil resistance and genetic polymorphism for CYP2C19 or its inhibition by other drugs would not affect efficacy.

In healthy Caucasians, the pharmacokinetics of proguanil, cycloguanil and atovaquone were not affected by combining the drugs. In patients with *P. falciparum* malaria, the
kinetics of proguanil given with atovaquone were comparable to those in healthy volunteers treated with proguanil only \[128\]. In a population pharmacokinetic study, there appeared to be a difference in the oral clearance of proguanil between poor and extensive metabolisers and a small but clinically insignificant difference between racially different groups comparable with proguanil monotherapy \[178\].

Inversely, the population kinetics of atovaquone as an object of interactions were also studied in a population of patients with falciparum malaria \[147\]. There was no effect of coadministration on the kinetics of atovaquone. Coadministration of pyrimethamine or tetracycline also had no significant effect on the disposition of atovaquone.

3.7.3 Pyrimethamine and Dapsone

This combination (Maloprim\textsuperscript{\textregistered}) is used mainly as a prophylactic in areas where chloroquine resistant is prevalent. At a high level of pyrimethamine resistance, the combination is probably not effective \[119\]. Despite extensive use, there is little pharmacokinetic information on this combination. In a multiple dose study, it was shown that the elimination half-lives and clearances were comparable to those found in single drug studies \[119\].

3.7.4 Combinations with Sulfonamides

The sulfonamides have all been used in combination with other agents, mainly other antifolates, but also in fixed combinations, for example with mefloquine. The main potential source of interaction with other drugs is high affinity for plasma proteins, especially of sulfadoxine, but this is unlikely to be clinically significant. Most fixed combinations with sulfonamides have stood the test of time, despite very little pharmacokinetic data to support the clinical experience. The most important fixed combination, sulfadoxine plus pyrimethamine, Fansidar\textsuperscript{\textregistered}, widely used as a first-line treatment of falciparum malaria in large areas of the world, is already facing its end because of resistance. Pharmacokinetic interactions between the 2 compounds have not been found.

4. Conclusion

The concept of combining drugs has become integrated into the evidence-guided approach to chemotherapy of infectious diseases. Combinations are important in the treatment of individual patients as well as in national policies concerning first-line drugs. When considering combination therapy, microbial resistance, the intrinsic antimicrobial activity of the respective agents and their synergy should be taken into account.
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Although the arguments for combination therapy are sound, they lead to considerable debate, not least because malaria affects large numbers of patients living mainly in areas where economic arguments strongly influence the cost-benefit balance [1]. Nevertheless, combination therapy is indispensable to combat multidrug-resistant parasites. Combination therapy has become the standard in the treatment of patients with infections such as tuberculosis, leprosy and HIV. For malaria, there is still a long way to go. In the majority of sub-Saharan nations, chloroquine is still the first-line drug for treatment of malaria and many fear that the change from chloroquine to sulfadoxine-pyrimethamine has come too late, knowing that resistance to the antifolates is already prominent.

Other combinations of drugs will be applied. Inevitably, the use of artemisinin drugs will increase because there is no known resistance to these drugs. Artemisinin drugs should be used in combination with other agents, and consequently interactions will increasingly become an important point of interest. In this review we have presented the currently available knowledge on interactions between antimalarial agents. Two distinct features have emerged. The first is that relatively little is known about pharmacokinetic interactions between antimalarial agents in humans. The magnitude of malaria as a global problem certainly merits more research into drug interactions, not only between antimalarial agents but also between several other groups of drugs, at least the agents on the essential drugs list. The second is that the limited information available suggests that interactions between antimalarial agents are rare. This experience is, for example, much different from that with the antiretroviral agents: pharmacotherapy of patients with HIV infection has become a labyrinth of interactions. Clearly, the short duration of antimalarial treatment courses is important here, but also the drugs themselves allow for many combinations without losing efficacy or increasing toxicity.

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References


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37. Karbwang J, Na Bangchang K. Clinical application of mefloquine pharmacokinetics in the

38. Na Bangchang K, Karbwang J, Back DJ. Mefloquine metabolism by human liver

Chongsuphajaisiddhi T, Farinotti R. Stereoselective pharmacokinetics of mefloquine

40. Hellgren U, Berggren-Palm e I, Bergqvist Y, Jerling M. Enantioselective pharmacokinetics

41. Looareessuwan S, White NJ, Warrell DA, Forgo I, Dubach UG, Ranalder, UB, Schwartz
DE. Studies of mefloquine bioavailability and kinetics using a stable isotope
technique: a comparison of Thai patients with falciparum malaria and healthy

42. Coleman MD, Fleckenstein L, Heiffer MH. Primaquine disposition in the isolated perfused
1989;10(2):153-64.

43. Leo KU, Wesche DL, Marino MT, Brewer TG. Mefloquine effect on disposition of


45. Milton KA, Edwards G, Ward SA, Orme L, Breckenridge AM. Pharmacokinetics of

46. ter Kuile FO, Dolan G, Nosten F, Edstein MD, Luxemburger C, Phaipun,
Chongsuphajaisiddhi T, Webster HK, White NJ. Halofantrine versus mefloquine in


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100. Murdoch RT, Ghabrial H, Mihaly GW, Morgan DJ, Smallwood RA. Malaria infection impairs glucuronidation and biliary excretion by the isolated perfused rat liver. Xenobiotica 1991; 21(12): 1571-82.


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Pharmacokinetic interactions of antimalarial agents


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