REVIEW

Artemisinins

C J Woodrow, R K Haynes, S Krishna

Postgrad Med J 2005;**81**:71–78. doi: 10.1136/pgmj.2004.028399

Artemisinins were discovered to be highly effective antimalarial drugs shortly after the isolation of the parent artemisinin in 1971 in China. These compounds combine potent, rapid antimalarial activity with a wide therapeutic index and an absence of clinically important resistance. Artemisinin containing regimens meet the urgent need to find effective treatments for multidrug resistant malaria and have recently been advocated for widespread deployment. Comparative trials of artesunate and quinine for severe malaria are in progress to see if the persistently high mortality of this condition can be reduced.

Artemisinins are derived from a plant called sweet wormwood (or sweet Annie: *Artemisia annua*). In China, where they were first discovered, ''*qinghao*'' extracts were reported to have antipyretic properties more than 1500 years ago. In 1967 an outstanding coordinated programme was started by the Chinese government to discover antimalarial principles in various medicinal herbs including *qinghao*. In 1971, a highly active chemical from *qinghao*, known as *qinghaosu* was obtained and is now called artemisinin.[1] Since this initial discovery, an array of semi-synthetic oil and water soluble derivatives of artemisinin have been developed, with a variety of formulations entering clinical studies. These compounds have impressive parasiticidal properties in vitro, rapidly arresting parasite metabolism in concentrations within the lower nanomolar range, and killing parasites more quickly than other antimalarial drugs.[2] These and other properties described below make artemisinins our most important class of antimalarial agent, and a mainstay against otherwise multidrug resistant *Plasmodium falciparum*. Their use in many countries has been severely restricted by cost, because artemisinins in combination are several-fold more expensive than the now almost useless chloroquine, or sulfadoxine-pyrimethamine, whose efficacy is also waning. However, providing mechanisms and the political will to subsidise and control the use of artemisinins can be implemented, it is probable that some regimens combining artemisinins with other antimalarials[3] will supersede cheaper and now ineffective alternatives.[4] Registration of artemisinins for use in developed countries is being actively pursued but only one fixed dose oral combination (artemether-lumefantrine) is so far available to treat uncomplicated malaria. If available, parenteral artemisinins can be used to treat severe malaria in the UK on a named patient basis.

See end of article for authors' affiliations

Correspondence to: Professor S Krishna, Department of Cellular and Molecular Medicine, Infectious Diseases, St George's Hospital Medical School, Cranmer Terrace, Tooting, London SW17 0RE, UK; s.krishna@sghms.ac.uk

Submitted
3 September 2004
Accepted
22 September 2004

CHEMISTRY AND SYNTHESIS

Artemisinin is comparatively easily purified by crystallisation after extraction from *Artemisia annua* plants but is extremely difficult to synthesise de novo. Artemisinin is a sesquiterpene lactone structure in which antimalarial activity is inextricably linked to an unusual endoperoxide trioxane moiety[5] (fig 1). Artemisinin itself is a highly crystalline compound that does not dissolve in oil or water and so can only be given by the enteral route. Artemisinin is the parent compound for semi-synthetic derivatives that have been chemically modified at the C10 position to produce artesunate, artemether, arteether, dihydroartemisinin, and artelinic acid (fig 1). These compounds have variously been formulated for oral, rectal, and parenteral administration. The sodium salts of artesunate and artelinate are used for parenteral administration of these derivatives.

Arteether was developed under the aegis of the World Health Organisation despite lacking clear advantages over artemether, for which a much larger clinical experience already exists; arteether is no longer being investigated as an antimalarial agent. However, locally formulated products are used in India (αβ arteether, E-mal) and the Netherlands (β-arteether, Artemotil (Artecef)). Artelinic acid (a water soluble derivative) was developed by Walter Read Army Institute for Research. Although artelinate will not be further developed, various formulations and combinations of artesunate with other antimalarials are under active development.

METABOLISM AND PHARMACOKINETICS

Once absorbed, the artemisinin derivatives are converted primarily to dihydroartemisinin (DHA) and thence to inactive metabolites via hepatic cytochrome P-450 and other enzyme systems.[5] DHA is itself a potent antimalarial with an elimination half life of about 45 minutes.[6 7] The extent of conversion to DHA differs between derivatives.[8] Artemisinin itself is not metabolised to DHA but acts as the primary antimalarial, while artesunate is rapidly (within minutes) hydrolysed to DHA and its antimalarial activity is largely mediated by DHA. Artemether and arteether contribute to antimalarial activity, probably to a similar extent as DHA, to which they are converted more slowly. DHA is mostly (90%) bound to plasma proteins.[9]

Pharmacokinetic studies on artemisinins have been limited by difficulties of assay; several techniques with differing accuracies have been used by various groups.[10] Furthermore, studies must necessarily take into account active metabolites (mostly DHA). Bioassay techniques measuring total antimalarial activity account for

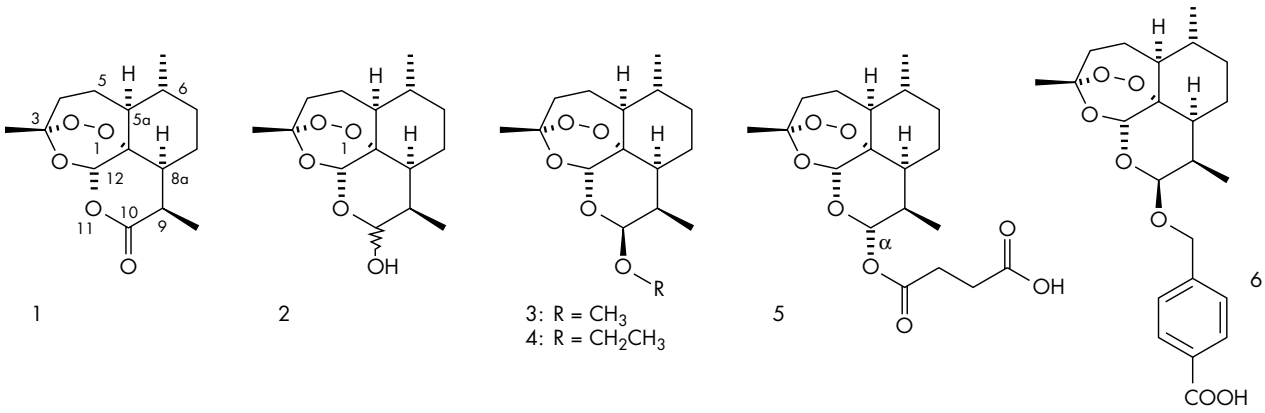

Figure 1 QTng hāo su or artemisinin 1 and derivatives dihydroartemisinin 2, artemether 3, arteether 4, artesunic acid (artesunate) 5, and artelinate 6. The numbering scheme is that used by Chemical Abstracts.

this[11] and, along with advances in assay methods, have allowed clearer pharmacokinetic profiling to emerge for drug formulations that have often been used empirically for many years. These studies are improving our understanding pharmacodynamic and toxicological aspects of this group of compounds.

In uncomplicated malaria, when artemisinins are used orally, most pharmacokinetic information is now available for artesunate followed by artemether. The absolute bioavailability of antimalarial activity after a single dose of oral artesunate in uncomplicated adult malaria is about 60%[6 12] although there is greater interpatient variation than in healthy volunteers.[13] Time to maximum DHA concentration is typically one to two hours.[6 13–18] Studies suggest that clearance after artesunate is reduced during acute infection compared with recovery, either via disease effects on pharmacokinetics or enzyme autoinduction.[12 13 19]

Although absolute bioavailability studies for artemether, artemisinin, and DHA are not possible given lack of intravenous formulations, pharmacodynamic activity (parasite clearance) after oral dosing of these derivatives is satisfactory. When studied, oral formulations show appropriately reliable and rapid absorption in the treatment of uncomplicated malaria.[20–22] As for artesunate, studies of oral artemether[23] and artemisinin[24 25] show increasing clearance with multiple dosing and during recovery from acute infection.

In severe malaria, the delayed and variable absorption of the oil soluble derivatives artemether and arteether when given by the intramuscular route is of great potential clinical relevance. Table 1 gives the pharmacokinetic data from studies on intramuscular artemether and artesunate in malaria patients.

In uncomplicated malaria, the relative bioavailability of intramuscular artemether appears poor compared with the oral route[20] and in most studies absorption is extremely variable with maximum concentrations only being achieved many hours after administration.[20 28 29] Most worryingly, a significant number of patients (5 of 26 in one study and 7 of 97 in another) had no detectable antimalarial present as ascertained by both conventional[28 29] and bioassay techniques[20 28] and this phenomenon was associated in one study with impaired parasite clearance kinetics.[28] These properties point to pharmacokinetic disadvantages for intramuscular artemether in severe malaria. Intramuscular arteether may be absorbed even more slowly than artemether.[30] These pharmacokinetic findings also have relevance to our understanding of neurotoxicity seen in animal models receiving oil based artemisinins (see below).

Parenteral artesunate is pharmacokinetically superior to artemether for the treatment of severe malaria, whether given intravenously[26 31] or by the intramuscular route (to children),[26] a fact that escaped attention in a later study on artemether.[29] Absorption from the intramuscular site in both adults with uncomplicated malaria and children with severe malaria is rapid with peak DHA concentrations achieved within one hour and DHA bioavailability over 80%[7 26] (table 1). Severity of malaria infection seems to have no significant influence on artesunate pharmacokinetics[31] but age may have.

Table 2 gives the pharmacokinetic data from studies on intrarectal administration of artemisinins to malaria patients.

Rectal artesunate in African children with moderate malaria (defined as being unable to take oral medications or prostration/obtundation) shows rapid but variable absorption with peak plasma DHA concentrations appearing in about two hours and bioavailability of between 20% and 60%.[34–36] Rectal artemisinin may have a comparatively slower absorption profile in volunteers and patients with uncomplicated malaria.[25 32 33 37] Intrarectal DHA has been studied in only a small number of patients and its behaviour seems comparable to intrarectal artemisinin.[7]

Unfortunately very few pharmacokinetic studies have focused on the variation in artemisinin profiles in different populations of patients, particularly children and pregnant women. There are also comparatively few studies of interactions between artemisinins and other antimalarial or groups of drugs, although there seem to be no significant interactions between artesunate and mefloquine or artemether and lumefantrine.[38]

ANTIMALARIAL PROPERTIES

Artemisinins kill all species of plasmodium that infect humans.[1 39–41] In vitro *P falciparum* IC_{50} values (median and range) have been reported as 4.2(0.5–34.6), 4.3(0.5–23.2), and 16.2(1.3–58.3)nM for artesunate, dihydroartemisinin, and artemether respectively.[42] The asexual stages of infection are the most susceptible, with artemisinins inducing up to a 10 000-fold reduction in parasite biomass per asexual cycle.[43] In common with other antimalarials, artemisinins are particularly active against the large ring stage of infection when parasites are beginning to become most metabolically active. However, in contrast with other currently useful antimalarials, artemisinins also target tiny ring stages of infection[44 45] (present only a few hours after red cells are invaded by merozoite stages). This killing results in removal of parasites from within infected cells, probably by the reticuloendothelial system, which returns these ''pitted'' erythrocytes to the

Table 1 Single dose conventional pharmacokinetic studies of intramuscular artemisinin derivatives given to patients with *Plasmodium falciparum* malaria*

Drug	Ref	Age (y)	Sev	Number	Dose	C_{max} μM	T_{max} (min)	$T_{1/2}$ (min)	$AUC_{0-\infty}$ (μM min)	V/F (l kg)	CL/F (l kg/h)	T_{abs}(min)
ARTS levels after ARTS												
	7	Adults	U	11	120 mg	2.3 (2.0–4.8)†	12 (10–15)†	41 (18)	156 (114)	2.6 (1.2)	2.9 (1.2)	
	26	1.5–10	S	14	1.2 mg/kg	1.6 (0.6–2.97)	7.2 (4.1–11.4)	25.2 (4.2–501)	83.5 (9.2–747.3)	1.3 (0.5–3.2)	2.4 (0.3–20.4)	2.7 (0.87–5.99)
				14	2.4 mg/kg	1.72 (0.7–10.1)	8 (3.9–78.9)	48.2 (13.4–319.7)	84.9 (39.2–2264.7)	2.1 (0.3–6.4)	3.48 (0.18–10.2)	2.5 (1.28–41.52)
DHA levels after ARTS												
	7	Adults	U	11	120 mg	4.1 (3.2–4.6)†	45 (34–60)†	64 (21)	522 (204)	1.1 (0.4)	0.73 (0.21)	
	26	1.5–10	S	14	1.2 mg/kg	1.2 (0.1–2.9)	25.9 (10.8–71.9)	31.9 (18.2–110.4)	83.6 (17.4–298.2)	1.2 (0.4–6.3)	2.16 (0.06–10.8)	15.4 (3.8–47.9)
				14	2.4 mg/kg	2.2 (0.8–3.6)	40.5 (11.5–68.2) (h)	40.2 (1.4–148.8) (h)	236.9 (46.7–582.7)	1.2 (0.03–3.2)	1.5 (0.36–7.8)	25.1 (5.1–47.3)
ARTM levels after ARTM												
	27	Adults	S	11	160 mg	0.84 (0.56–1.81)	4 (2–6)	5.7 (4.2–6.6)	811 (431–1702)	8.6 (4.2–12.3)	1.1 (0.5–1.5)	
		Adults	S+RF	6	160 mg	1.28 (1.01–2.12)	4 (2–6)	7.0 (5.5–10)	1139 (708–1702)	5.4 (3.2–6.9)	0.4 (0.3–0.8)	
DHA levels after ARTM												
	27	Adults	S	11	160 mg	1.28 (0.89–1.94)	4 (4–6)		898 (435–1806)			
		Adults	S+RF	6	160 mg	1.42 (1.09–2.35)	4 (4–4)		1063 (756–1806)			
Bioassay after ARTM												
	20	Adults	U	16	2 mg/kg	0.121 (.02–0.37)	8 (4–24)		163 (33–962)		2.46 (0.48–12.03)	

*Data are presented as median (range) or mean (SD) unless otherwise indicated. Bioassay values are DHA equivalents. ARTS, artesunate; DHA, dihydroartemisinin; ARTM, artemether; U, uncomplicated malaria; S, severe malaria; RF, acute renal failure; C_{max}, peak drug concentration in plasma; T_{max}, observed time to C_{max}; $T_{1/2}$, elimination half life; AUC, area under the plasma concentration-time curve; V/F, fractional volume of the central compartment; CL/F, fractional clearance; T_{abs}, absorption half time. †Interquartile range.

Table 2 Single dose pharmacokinetic studies of intrarectal artemisinin derivatives administered to patients with *Plasmodium falciparum* malaria*

Drug	Ref	Age group	Severity	Number	Dose	C_{max} μM	T_{max} (h)	$T_{1/2}$ (h)	AUC (μM min)	V/F (l kg)	CL/F (l kg/h)	T_{abs}(h)	T_{lag}(h)	B (%)
ARN after ARN														
	32	Adults	U	8	600 mg	0.37 (0.21)	7.2 (3.9)	3.1 (2.1)		80.8 (61.9)				
	25	Adults	U	15	500 mg	0.66 (0.33)	4.0 (2–10)	2.0 (1.4)	184 (110)¶				1 (0.5–2.5)	
	33	Adults	U	8	600 mg	0.47 (0.1–0.6)	6.5 (2–14)		257 (26–406)¶				0.3 (0.3–0.3)	
DHA after DHA														
	7	Adults	U	11	160 mg	0.75 (0.55–1.11)†	4.0 (2.63–6)†		204 (78)‡					16 (13–25)†
ARTS after ARTS														
	34**	5–10	U††	47	12.7 (0.9) mg/kg	1.085‡‡	0.9‡‡	0.27 (0.13)				2.3 (1.0)		
DHA after ARTS														
	35	1.5–7	M	10	9.3 mg/kg (6.9–11.8)	2.4 (0.8–5.8)	1.7 (0.9–3.2)	0.79 (0.41–2.69)	588 (84–1692)¶	4.4 (1.8–14.4)	2.6 (1–22.3)	0.69 (0.3–1.24)	0.63 (0–1.36)	58 (24–131)
				16	18.9 mg/kg (15.4–22.9)	3.1 (0.7–6.8)	1.8 (0.6–3.3)	0.85 (0.09–2.5)	792 (174–1572)¶	5.9 (1.1–11.7)	3.9 (1.7–19.6)	1.1 (0.6–2.7)	0.37 (0–0.93)	23 (6–78)
	34**	5–10	U††	47	12.7 (0.9) mg/kg	2.52‡‡	2.3‡‡	0.71 (0.22)		2.15‡‡	2.25‡‡			

*Data are presented as median (range) or mean (SD) unless otherwise indicated. ARN, artemisinin; DHA, dihydroartemisinin; ARTS, artesunate; U, uncomplicated malaria; M, moderate malaria; C_{max}, peak drug concentration in plasma; T_{max}, observed time to C_{max}; $T_{1/2}$, elimination half life; AUC, area under the plasma concentration-time curve; V/F, fractional volume of the central compartment; CL/F, fractional clearance; T_{abs}, absorption half time; T_{lag}, time to quantifiable drug in plasma; B, relative bioavailability. †Interquartile range, ‡0–8, ¶0-∞, **population pharmacokinetic study, ††includes patients with *P vivax* infection, ‡‡modelled mean values.

circulation carrying an immunological marker of the presence of the parasite on its surface (an early stage antigen called RESA).[45] Artemisinins also inhibit metabolism of parasites more quickly than other antimalarials used to treat severe malaria,[44 46] a pharmacodynamic property that is of potential benefit given that most deaths in African children occur in the first 12 to 24 hours after admission. They also reduce cytoadherence of infected red cells, a recognised virulence determinant.[47]

Artemisinins do not interfere with hepatic stages of parasite development and therefore have no causal prophylactic value. They do kill early gametocyte stages of development and have the potential to interfere with mosquito transmission.[48] This property may be useful in areas where transmission rates for malaria are comparatively low,[49] but has not provided benefit in areas of high transmission despite reported reduction of gametocyte rates.[50 51]

MECHANISM OF ACTION

For several decades, the antimalarial action of artemisinins has been attributed to their chemical capability to generate free radicals. This mechanism of action has been suggested partly on the grounds that well recognised sources of free radicals (such as *tert*-butylperoxide) can themselves kill malaria parasites, albeit in comparatively high (mM) concentrations.[52] The peroxide structure (essential for antimalarial activity[53 54]) has been studied in detailed chemical experiments aiming to decipher exactly how it may act as an antimalarial. It is held by many workers that artemisinins upon reaction with Fe^{2+} are converted first into oxygen centred free radicals derived by reductive cleavage of the peroxide bridge, which are then converted into carbon centred free radicals by intramolecular hydrogen abstraction from CH_2 groups on the periphery of the artemisinin by the O centred radicals. Fe^{2+} is a catalyst that can generate free radicals from peroxidic structures in other peroxides, but in the case of the antimalarial action or artemisinins, this is further maintained to take place in the food vacuole by either free Fe^{2+} or by ferroprotoporphyrin IX (reduced haem).[55] Carbon centred free radicals have been put forward as principal intermediates in the parasiticidal process, but this theory of action sees artemisinins killing parasites via an indiscriminate process, a view that is hard to integrate with the exceptionally high in vitro activities of artemisinins and stands in pronounced contrast with the mechanism of action of most bioactive molecules where activity is mediated by high affinity binding to an active site.

More recently, an alternative mechanism of action for artemisinins based on inhibition of the malarial parasite's calcium ATPase (sarcoplasmic endoplasmic reticulum calcium ATPase, SERCA) has been suggested.[56] This work has reconciled some intriguing observations on actions of artemisinins, and also proposed new directions for further studies and drug development pathways. The arguments for and against these different mechanisms have been discussed in detail in current reviews.[53 54 57]

CLINICAL APPLICATIONS

Artemisinin derivatives are used for treatment of uncomplicated and severe malaria in both adults and children. After some initial concerns, evidence for the safety of artemisinins in pregnant women (a population that is particularly at risk from malaria) is emerging; in a study of over 500 women treated with artemisinins in Thailand, there was no increase in rate of abortion, congenital abnormality, or stillbirth compared with background incidences in this population.[58] When artesunate was added to an intermittent pyrimethamine-sulfadoxine regimen in pregnant women in the Gambia, there was again no significant adverse effect after gestational exposure.[59] However, data on artemisinin use in the first trimester of pregnancy remain scanty, and more experience is needed before recommendations can be made on a firm basis.

Artemisinins are unique among antimalarials in that there is still no evidence of significant resistance in clinical isolates. Their short half life renders them inappropriate for prophylaxis.

Uncomplicated malaria

Uncomplicated malaria can be managed by oral antimalarial and symptomatic therapy, in contrast with moderate or severe disease. Particular combinations have been reviewed recently.[3] The emergence of resistance to chloroquine and pyrimethamine-sulfadoxine has led to the introduction of artemisinin containing combinations, particularly in south east Asia where resistance to mefloquine also emerged rapidly.[60] In this location combination of artemisinins with mefloquine provided much improved cure rates.[61 62] Successful use of artemisinin derivatives with sulfadoxine-pyrimethamine has recently been described in Africa[63] but addition of artesunate to chloroquine did not prevent treatment failure.[64] The combination approach has been discussed extensively elsewhere[3 65 66] and is now being implemented in a variety of national policies as well as by international organisations. However, with ready availability on the open market, the reality is that artemisinins are certainly being applied in inappropriate regimens including monotherapy (see below); furthermore trading of fake artesunate represents a significant threat to malaria initiatives.[67] Artemether-lumefantrine is the only fixed dose artemisinin containing combination that is registered for use in Europe, and is licensed as a six dose regimen over 60 hours in patients weighing over 35 kg.[68]

Very few dose ranging/frequency studies have been carried out to ensure that current regimens for uncomplicated malaria have been truly optimised. In adults, different doses of artesunate, given under cover of the slower acting agent mefloquine, suggested to the authors that a dose of 2 mg/kg artesunate was sufficient to reduce parasitaemia rapidly.[69] However, results from only two to three patients probably skewed the inherently variable pharmacodynamic data obtained in this small study, and make more tentative the conclusions drawn from this study about oral dosing regimens in general. Most physicians currently use an oral dose of artesunate of 4 mg/kg per day for three days for patients with uncomplicated malaria when in combination with a second antimalarial. Despite the generally rapid elimination kinetics of artemisinins, daily dosing of oral artesunate results in parasite clearance kinetics indistinguishable from twice daily dosing.[61] This suggests that constant drug levels are not necessary for satisfactory parasite clearance; the biological effects of artemisinins extend beyond their presence at therapeutic concentrations in plasma,[43] in some ways analogous to a post-antibiotic effect.

Severe malaria

Severe malaria in hospitalised patients is associated with a mortality of between 15% to 20%, despite appropriate antimalarial and supportive treatment.[70] With the widespread establishment of chloroquine resistance, there are only two classes of compound that are useful to manage severe malaria, the cinchona alkaloids (quinine and quinidine) and artemisinins. In Europe and Africa, quinine remains the drug of choice, although it suffers from certain drawbacks. Quinine has a narrow therapeutic ratio, causing hyperinsulinaemic hypoglycaemia (more frequent and severe in pregnancy) and prolongs the QT_c interval when given parenterally, particularly if infused too rapidly. Intramuscular

quinine is effective, but can cause local toxicity as well as hypoglycaemia in patients who may not have intravenous access. Furthermore, in south east Asia there is evidence of increasing quinine resistance[71] so that artemisinins are now used as first line treatment for severe malaria in most units.

Several trials have compared quinine and intramuscular artemether for severe infection in both south east Asia[72 73] and Africa.[74–78] Despite improved parasite clearance parameters in most trials, definitive evidence for improved mortality with artemether in individual trials and meta-analysis is lacking.[79–81] Many important observations have emerged from these studies. Firstly, the incidence of post-admission hypoglycaemia is significantly higher with quinine compared with artemether.[72] Secondly, the frequency of dosing (more with quinine) also adds extra demands on scarce nursing resources. Most significantly, artemether may not have been the best choice of artemisinin to study in the first place, as suggested more than a decade ago (Dr Hien, Cho Quan Hospital, Vietnam, personal communication to SK). Compared with artesunate, artemether is less completely biotransformed to the more potent dihydroartemisinin and has slow, erratic absorption after intramuscular administration (see above); in fact the ability of artemether to provide equivalent benefit to quinine is probably testament to the antimalarial potency of the artemisinin derivatives as a group.

Attention has therefore switched to artesunate. Parenteral artesunate has been used in adults and children with severe malaria in south east Asia[82–84] where intramuscular administration was comparable in efficacy and safety to the intravenous route.[82 84] In an analogous manner to parenteral artemether, artesunate (intravenously) shows reduced incidence of hypoglycaemia compared with quinine.[85] Large multinational studies in south east Asia comparing artesunate and quinine using mortality as an end point are now underway (Professor N White, personal communication). Similar studies in African children are also urgently needed because of differences in natural history of severe malaria, particularly the more rapid recovery of children compared with adults as well as the incidence of quinine resistance in south east Asia, both of which may obscure mortality advantages seen with quinine in adults. Intramuscular artesunate has an acceptable pharmacokinetic profile in African children[26] where parasite clearance kinetics seem to be comparable to the intravenous route. Trials in this area are a high priority and can properly be funded by organisations such as the EDCTP and Medicines for Malaria Ventures whose avowed aim is to improve treatments for malaria.

Intrarectal administration

Patients with malaria presenting in rural areas may be obtunded or vomiting and unable to take oral medications, leading to significant delay in treatment if facilities for parenteral treatment are unavailable. In such circumstances the rectal route of administration is attractive because in areas where this route is culturally acceptable, rural healthcare workers can be trained to identify moderate and severe malaria and administer rectal drugs before transfer of patients to hospital. Quinine has been tested via the intrarectal route[86] but may still induce hypoglycaemia, which may not be recognised or treated. The wider therapeutic index of artemisinin derivatives means that they are excellent choices for rectal administration despite the inevitable variability of absorption from this route. Artemisinin formulations have been used with empirical success in south east Asia for some considerable time[87] and recently pharmacokinetic profiles have begun to be delineated.[25 33 35 36 88] In a comparative study with parenteral quinine, rectal artesunate was efficacious in African children with moderate malaria.[89] This study was developed from detailed pharmacokinetic characterisation of a rectal formulation of artesunate that led to rapid falls in parasitaemia that were indistinguishable from those seen after intravenous artesunate.[35]

LIMITATIONS OF ARTEMISININS

Putting aside questions of cost, which may be the most important for users of antimalarials but have been comprehensively reviewed in a recent authoritative report from the Institute of Medicine,[4] there are certain inherent problems with current artemisinins that require discussion.

Poor cure rate of monotherapy

Artemisinins reliably reduce initial malaria parasitaemia by a factor of 10^4 per 48 hour asexual cycle and modelling studies therefore suggest that six days of treatment should cure parasite burdens of up to 10^{12} parasites. This model is difficult to reconcile with the high recrudescence rates (10%–15%) seen with artemisinin monotherapy. This poor efficacy of cure (which is not due to resistance) is usually attributed to the intrinsically short half life of artemisinins, which is further shortened by the increased drug clearance that develops during repeat dosing and/or convalescence with various oral artemisinin derivatives (see above).[12 13 19 23–25] Blaming pharmacokinetic factors alone for the poor efficacy of artemisinin monotherapy may not be justified because constant drug levels are not necessary for potent pharmacodynamic effects (at least in the initial, visible phase of parasitaemia). Furthermore, if pharmacokinetic behaviour were a problem, prolongation of treatment course may be predicted to compensate,[43] but this is not generally observed in practice[90 91]; seven days of monotherapy with artemisinin still only cures 80%–90% of uncomplicated falciparum infections. Parasite reduction ratio models for artesunate derived on data obtained at the start of treatment may not be applicable to the process of eradication of small numbers of residual parasites, which determines eventual cure rates.[43] Other phenomena may exist that permit escape from artemisinin therapy, necessitating a second (albeit less visibly effective) antimalarial.[43] Although it has been strongly argued that, in any case, combination therapy has long term benefit in preventing resistance,[92 93] the poor efficacy of monotherapy with the current generation of artemisinins remains a troubling and poorly explained phenomenon.

Neurotoxicity

Despite pre-clinical evidence of brainstem toxicity in animals,[94 95] millions of doses in various formulations have been given to humans without significant evidence of major toxicity, even when particular attention is given to monitoring for

Box 1 Pharmacokinetics of artemisinin derivatives

- Oral formulations of artemisinin derivatives are generally rapidly absorbed.
- Intramuscular artemether has been used in many studies on severe malaria, but has slow and erratic absorption.
- Intramusucular artesunate is pharmacokinetically superior to artemether for the treatment of severe malaria, showing rapid and reliable absorption.
- Intrarectal artesunate shows rapid absorption and is a promising treatment for patients with moderate malaria when oral administration is not possible, and until hospital care is available.

Box 2 Key references

Eckstein-Ludwig U, Webb R, Van Goethem ID, *et al*. Artemisinins target the SERCA of Plasmodium falciparum. *Nature* 2003;**424**:957–61.
Artemether-Quinine Meta-analysis Study Group. A meta-analysis using individual patient data of trials comparing artemether with quinine in the treatment of severe falciparum malaria. *Trans R Soc Trop Med Hyg* 2001;**95**:637–50.
Giao PT, Binh TQ, Kager PA, *et al*. Artemisinin for treatment of uncomplicated falciparum malaria: is there a place for monotherapy? *Am J Trop Med Hyg* 2001;**65**:690–5.
White NJ. Antimalarial drug resistance. *J Clin Invest* 2004;**113**:1084–92.
Gordi T, Lepist EI. Artemisinin derivatives: toxic for laboratory animals, safe for humans? *Toxicol Lett* 2004;**147**:99–107.

neurotoxicity both clinically[96 97] and pathologically.[98] This discrepancy between animal and human toxicity has been attributed to the comparatively high and prolonged dosing regimens used in certain animal studies.[99] In addition, pharmacokinetic studies of parenteral artemether and arteether have showed the slow release and consequently long exposure times seen with oil based formulations of these drugs in both animals[100] and humans.[20 30 101] It is probably the duration of exposure to artemisinins that determines neurotoxicity rather than the maximum concentrations reached.[102] Prolonged high concentrations of artemisinins are certainly not seen in oral regimens, which constitute the vast majority of artemisinin courses given, and there is no pathological evidence of neurotoxicity in patients exposed to an average of 76.5 hours of intramuscular artemether.[98] A recent claim that artemether-lumefantrine induces mild but significant hearing loss[103] seems to contradict this view but needs to be reproduced independently and the mechanism dissected, particularly in terms of the time course of hearing loss.[99] Concern with regard to neurotoxicity should also be maintained in the context of children who have more vulnerable neurological systems and where therapeutic experience is more limited. Even taking into account these concerns, artemisinin derivatives have less major toxicity than other available antimalarial drugs.

Other toxicity and interactions

Administration of artemisinins may be associated with transient gastrointestinal disturbance, a characteristic of acute malaria in any case, and rarely with severe allergic reactions[104] or haemolysis.[105] Fetotoxicity is an important concern, again based on animal studies, although artemisinins have not been shown to be teratogenic in the small human experience available. They are not advised for use in the first trimester of pregnancy, but have been used rarely when alternatives to lifesaving treatment have been exhausted. Given the plan to roll out artemisinin combination therapies, there have been few drug metabolism and interaction studies carried out for artemisinins and their combination partners.[10] In addition, there are few stability studies for many of the formulations of artemisinins (mainly artesunate) that are used today.

ARTEMISININS—THE NEXT GENERATION

Some limitations of current artemisinins may be addressed by well designed studies using available formulations of drugs. However, some issues may best be dealt with by developing the next generation of artemisinins, aiming for increased potency, reduced toxicity, and improved stability. In this regard, fully synthetic trioxalones under drug development may help rapidly to expand the repertoire of new antimalarials.[106] They have the advantage of independence from artemisinin as a raw material for synthesis. On the other hand newer semi-synthetic artemisinins such as artemisone (http://www.mmv.org/) have been developed from a much larger base of medicinal chemistry and clinical experience suggesting that both approaches to improving our stock of antimalarials should be pursued.

USES OUTSIDE MALARIA

Oral artemether has been known for some time to possess activity against immature worms of *Schistosoma japonicum* and *Schistosoma mansoni*, and has proved an efficacious chemoprophylactic agent against both infections.[107 108] It should be noted that the long term consequences of artemether use in this context potentially include selection for resistant plasmodia. Artesunate shows antitumour cell activity,[109] although it has yet to enter clinical trials.

CONCLUSION

Like many drugs, artemisinins have been used empirically for many years during which their mechanism of action and pharmacokinetic properties have been unclear. Empirical judgements of efficacy and optimal administration have tended to be influenced by their undoubtedly impressive parasite clearance kinetics, which are superior to other commonly used antimalarials.[110] However if the only fundamental and reliable measures of efficacy are cure and mortality rates for uncomplicated and severe malaria respectively, current artemisinins have some way to go before they can be said to provide a clear cut advantage over other antimalarial combinations in some geographical locations. Artemisinins are poorly efficacious at curing malaria as monotherapy, a phenomenon that is not well understood. Some concerns over neurotoxicity and its mechanism also remain. No regimen has yet proved superior to quinine for reducing mortality of severe malaria, although artemisinins certainly reduce the incidence of hypoglycaemia. Despite these issues, no time should be wasted in deploying artemisinins as part of combination therapy for multidrug resistant malaria when judged appropriate, with rectal administration permitting community based treatment of moderate malaria. If trial evidence can be obtained for improved outcome compared with quinine, parenteral artesunate may finally take its place as the optimum treatment for the ever present problem of severe malaria.

YES/NO QUESTIONS (ANSWERS AT END OF REFERENCES)

1. True or false:

(A) Artemisinins have very high efficacy in terms of cure rate when administered as monotherapy
(B) There are several independent reports of human brainstem neurotoxocity induced by artemisinins
(C) Parenteral artemisinins have been definitively shown to reduce mortality in severe malaria compared with quinine
(D) Oral artemisinins are not used in prophylactic regimens
(E) Artemisinin resistance has developed rapidly in south east Asia in the past decade
(F) Manufacture of current artemisinin formulations is now entirely synthetic

2. Which formulations of artemisinin show most promise in the context of severe malaria?

Authors' affiliations

C J Woodrow, S Krishna, Department of Cellular and Molecular Medicine, Infectious Diseases, St George's Hospital Medical School, Tooting, London, UK
R K Haynes, Department of Chemistry, The Hong Kong University of Science and Technology, Clear Water Bay, Kowloon, Hong Kong

Funding: none.

Conflicts of interest: none declared.

REFERENCES

1 **Qinghaosu Antimalaria Coordinating Research Group**. Antimalaria studies on Qinghaosu. *Chin Med J (Engl)* 1979;**92**:811–16.
2 **White NJ**. Clinical pharmacokinetics and pharmacodynamics of artemisinin and derivatives. *Trans R Soc Trop Med Hyg* 1994;**88**(suppl 1):S41–3.
3 **Kremsner PG**, Krishna S. Antimalarial combinations. *Lancet* 2004;**364**:285–94.
4 **Arrow KJ**, Panosian C, Gelband H. *Saving lives, buying time: economics of malaria drugs in an age of resistance*. Washington DC: National Academic Press, 2004.
5 **Haynes RK**. Artemisinin and derivatives: the future for malaria treatment? *Curr Opin Infect Dis* 2001;**14**:719–26.
6 **Batty KT**, Thu LT, Davis TM, *et al.* A pharmacokinetic and pharmacodynamic study of intravenous vs oral artesunate in uncomplicated falciparum malaria. *Br J Clin Pharmacol* 1998;**45**:123–9.
7 **Ilett KF**, Batty KT, Powell SM, *et al.* The pharmacokinetic properties of intramuscular artesunate and rectal dihydroartemisinin in uncomplicated falciparum malaria. *Br J Clin Pharmacol* 2002;**53**:23–30.
8 **Li QG**, Peggins JO, Fleckenstein LL, *et al.* The pharmacokinetics and bioavailability of dihydroartemisinin, arteether, artemether, artesunic acid and artelinic acid in rats. *J Pharm Pharmacol* 1998;**50**:173–82.
9 **Batty KT**, Ilett KF, Davis TM. Protein binding and alpha : beta anomer ratio of dihydroartemisinin in vivo. *Br J Clin Pharmacol* 2004;**57**:529–33.
10 **Navaratnam V**, Mansor SM, Sit NW, *et al.* Pharmacokinetics of artemisinin-type compounds. *Clin Pharmacokinet* 2000;**39**:255–70.
11 **Teja-Isavadharm P**, Peggins JO, Brewer TG, *et al.* Plasmodium falciparum-based bioassay for measurement of artemisinin derivatives in plasma or serum. *Antimicrob Agents Chemother* 2004;**48**:954–60.
12 **Newton P**, Suputtamongkol Y, Teja-Isavadharm P, *et al.* Antimalarial bioavailability and disposition of artesunate in acute falciparum malaria. *Antimicrob Agents Chemother* 2000;**44**:972–7.
13 **Teja-Isavadharm P**, Watt G, Eamsila C, *et al.* Comparative pharmacokinetics and effect kinetics of orally administered artesunate in healthy volunteers and patients with uncomplicated falciparum malaria. *Am J Trop Med Hyg* 2001;**65**:717–21.
14 **Benakis A**, Paris M, Loutan L, *et al.* Pharmacokinetics of artemisinin and artesunate after oral administration in healthy volunteers. *Am J Trop Med Hyg* 1997;**56**:17–23.
15 **Bethell DB**, Teja-Isavadharm P, Cao XT, *et al.* Pharmacokinetics of oral artesunate in children with moderately severe Plasmodium falciparum malaria. *Trans R Soc Trop Med Hyg* 1997;**91**:195–8.
16 **Na-Bangchang K**, Karbwang J, Congpoung K, *et al.* Pharmacokinetic and bioequivalence evaluation of two generic formulations of oral artesunate. *Eur J Clin Pharmacol* 1998;**53**:375–6.
17 **Binh TQ**, Ilett KF, Batty KT, *et al.* Oral bioavailability of dihydroartemisinin in Vietnamese volunteers and in patients with falciparum malaria. *Br J Clin Pharmacol* 2001;**51**:541–6.
18 **Suputtamongkol Y**, Newton PN, Angus B, *et al.* A comparison of oral artesunate and artemether antimalarial bioactivities in acute falciparum malaria. *Br J Clin Pharmacol* 2001;**52**:655–61.
19 **Khanh NX**, de Vries PJ, Ha LD, van Boxtel CJ, *et al.* Declining concentrations of dihydroartemisinin in plasma during 5-day oral treatment with artesunate for Falciparum malaria. *Antimicrob Agents Chemother* 1999;**43**:690–2.
20 **Silamut K**, Newton PN, Teja-Isavadharm P, *et al.* Artemether bioavailability after oral or intramuscular administration in uncomplicated falciparum malaria. *Antimicrob Agents Chemother* 2003;**47**:3795–8.
21 **De Vries PJ**, Tran KD, Nguyen XK, *et al.* The pharmacokinetics of a single dose of artemisinin in patients with uncomplicated falciparum malaria. *Am J Trop Med Hyg* 1997;**56**:503–7.
22 **Newton PN**, van Vugt M, Teja-Isavadharm P, *et al.* Comparison of oral artesunate and dihydroartemisinin antimalarial bioavailabilities in acute falciparum malaria. *Antimicrob Agents Chemother* 2002;**46**:1125–7.
23 **van Agtmael MA**, Cheng-Qi S, Qing JX, *et al.* Multiple dose pharmacokinetics of artemether in Chinese patients with uncomplicated falciparum malaria. *Int J Antimicrob Agents* 1999;**12**:151–8.
24 **Alin MH**, Ashton M, Kihamia CM, *et al.* Multiple dose pharmacokinetics of oral artemisinin and comparison of its efficacy with that of oral artesunate in falciparum malaria patients. *Trans R Soc Trop Med Hyg* 1996;**90**:61–5.
25 **Ashton M**, Nguyen DS, Nguyen VH, *et al.* Artemisinin kinetics and dynamics during oral and rectal treatment of uncomplicated malaria. *Clin Pharmacol Ther* 1998;**63**:482–93.
26 **Nealon C**, Dzeing A, Muller-Romer U, *et al.* Intramuscular bioavailability and clinical efficacy of artesunate in Gabonese children with severe malaria. *Antimicrob Agents Chemother* 2002;**46**:3933–9.
27 **Karbwang J**, Na-Bangchang K, Tin T, *et al.* Pharmacokinetics of intramuscular artemether in patients with severe falciparum malaria with or without acute renal failure. *Br J Clin Pharmacol* 1998;**45**:597–600.
28 **Murphy SA**, Mberu E, Muhia D, *et al.* The disposition of intramuscular artemether in children with cerebral malaria; a preliminary study. *Trans R Soc Trop Med Hyg* 1997;**91**:331–4.
29 **Mithwani S**, Aarons L, Kokwaro GO, *et al.* Population pharmacokinetics of artemether and dihydroartemisinin following single intramuscular dosing of artemether in African children with severe falciparum malaria. *Br J Clin Pharmacol* 2004;**57**:146–52.
30 **Looareesuwan S**, Oosterhuis B, Schilizzi BM, *et al.* Dose-finding and efficacy study for i.m. artemotil (beta-arteether) and comparison with i.m. artemether in acute uncomplicated P. falciparum malaria. *Br J Clin Pharmacol* 2002;**53**:492–500.
31 **Davis TM**, Phuong HL, Ilett KF, *et al.* Pharmacokinetics and pharmacodynamics of intravenous artesunate in severe falciparum malaria. *Antimicrob Agents Chemother* 2001;**45**:181–6.
32 **van Boxtel CJ**, van Agtmael MA, De Vries PJ, *et al.* Some pharmacokinetic and dynamic comparisons of artemisinin derivatives in man. *Jpn J Trop Med Hyg* 1996;**24**:49–54.
33 **Koopmans R**, Duc DD, Kager PA, *et al.* The pharmacokinetics of artemisinin suppositories in Vietnamese patients with malaria. *Trans R Soc Trop Med Hyg* 1998;**92**:434–6.
34 **Karunajeewa HA**, Ilett KF, Dufall K, *et al.* Disposition of artesunate and dihydroartemisinin after administration of artesunate suppositories in children from Papua New Guinea with uncomplicated malaria. *Antimicrob Agents Chemother* 2004;**48**:2966–72.
35 **Krishna S**, Planche T, Agbenyega T, *et al.* Bioavailability and preliminary clinical efficacy of intrarectal artesunate in Ghanaian children with moderate malaria. *Antimicrob Agents Chemother* 2001;**45**:509–16.
36 **Halpaap B**, Ndjave M, Paris M, *et al.* Plasma levels of artesunate and dihydroartemisinin in children with Plasmodium falciparum malaria in Gabon after administration of 50-milligram artesunate suppositories. *Am J Trop Med Hyg* 1998;**58**:365–8.
37 **Koopmans R**, Ha LD, Duc DD, *et al.* The pharmacokinetics of artemisinin after administration of two different suppositories to healthy Vietnamese subjects. *Am J Trop Med Hyg* 1999;**60**:244–7.
38 **Giao PT**, de Vries PJ. Pharmacokinetic interactions of antimalarial agents. *Clin Pharmacokinet* 2001;**40**:343–73.
39 **Li GQ**, Guo XB, Fu LC, *et al.* Clinical trials of artemisinin and its derivatives in the treatment of malaria in China. *Trans R Soc Trop Med Hyg* 1994;**88**(suppl 1):S5–6.
40 **Borrmann S**, Szlezak N, Binder RK, *et al.* Evidence for the efficacy of artesunate in asymptomatic Plasmodium malariae infections. *J Antimicrob Chemother* 2002;**50**:751–4.
41 **Same-Ekobo A**, Lohoue J, Essono E, *et al.* [Rapid resolution of Plasmodium ovale malarial attacks using artesunate (Arsumax)]. *Med Trop (Mars)* 1999;**59**:43–5.
42 **Brockman A**, Price RN, van Vugt M, *et al.* Plasmodium falciparum antimalarial drug susceptibility on the north-western border of Thailand during five years of extensive use of artesunate-mefloquine. *Trans R Soc Trop Med Hyg* 2000;**94**:537–44.
43 **White NJ**. Assessment of the pharmacodynamic properties of antimalarial drugs in vivo. *Antimicrob Agents Chemother* 1997;**41**:1413–22.
44 **ter Kuile F**, White NJ, Holloway P, *et al.* Plasmodium falciparum: in vitro studies of the pharmacodynamic properties of drugs used for the treatment of severe malaria. *Exp Parasitol* 1993;**76**:85–95.
45 **Angus BJ**, Chotivanich K, Udomsangpetch R, *et al.* In vivo removal of malaria parasites from red blood cells without their destruction in acute falciparum malaria. *Blood* 1997;**90**:2037–40.
46 **Hien TT**, White NJ. Qinghaosu. *Lancet* 1993;**341**:603–8.
47 **Udomsangpetch R**, Pipitaporn B, Krishna S, *et al.* Antimalarial drugs reduce cytoadherence and rosetting of Plasmodium falciparum. *J Infect Dis* 1996;**173**:691–8.
48 **Kumar N**, Zheng H. Stage-specific gametocytocidal effect in vitro of the antimalaria drug qinghaosu on Plasmodium falciparum. *Parasitol Res* 1990;**76**:214–18.
49 **Price RN**, Nosten F, Luxemburger C, *et al.* Effects of artemisinin derivatives on malaria transmissibility. *Lancet* 1996;**347**:1654–8.
50 **von Seidlein L**, Milligan P, Pinder M, *et al.* Efficacy of artesunate plus pyrimethamine-sulphadoxine for uncomplicated malaria in Gambian children: a double-blind, randomised, controlled trial. *Lancet* 2000;**355**:352–7.
51 **von Seidlein L**, Walraven G, Milligan PJ, *et al.* The effect of mass administration of sulfadoxine-pyrimethamine combined with artesunate on malaria incidence: a double-blind, community-randomized, placebo-controlled trial in the Gambia. *Trans R Soc Trop Med Hyg* 2003;**97**:217–25.
52 **Clark IA**, Hunt NH, Cowden WB, *et al.* Radical-mediated damage to parasites and erythrocytes in Plasmodium vinckei infected mice after injection of t-butyl hydroperoxide. *Clin Exp Immunol* 1984;**56**:524–30.
53 **Krishna S**, Uhlemann A-C, Haynes RK. Artemisinins: mechanisms of action and potential for resistance. *Drug Resistance Updates* 2004;**7**:233–44.
54 **Haynes RK**, Krishna S. Artemisinins: activities and actions. *Microbes Infect* 2004;**6**:1339–46.
55 **Meshnick SR**, Thomas A, Ranz A, *et al.* Artemisinin (qinghaosu): the role of intracellular hemin in its mechanism of antimalarial action. *Mol Biochem Parasitol* 1991;**49**:181–9.
56 **Eckstein-Ludwig U**, Webb R, Van Goethem ID, *et al.* Artemisinins target the SERCA of Plasmodium falciparum. *Nature* 2003;**424**:957–61.
57 **O'Neill PM**, Posner GH. A medicinal chemistry perspective on artemisinin and related endoperoxides. *J Med Chem* 2004;**47**:2945–64.
58 **McGready R**, Cho T, Keo NK, *et al.* Artemisinin antimalarials in pregnancy: a prospective treatment study of 539 episodes of multidrug-resistant Plasmodium falciparum. *Clin Infect Dis* 2001;**33**:2009–16.

59 **Deen JL**, von Seidlein L, Pinder M, *et al*. The safety of the combination artesunate and pyrimethamine-sulfadoxine given during pregnancy. *Trans R Soc Trop Med Hyg* 2001;**95**:424–8.
60 **Nosten F**, ter Kuile F, Chongsuphajaisiddhi T, *et al*. Mefloquine-resistant falciparum malaria on the Thai-Burmese border. *Lancet* 1991;**337**:1140–3.
61 **Nosten F**, Luxemburger C, ter Kuile FO, *et al*. Treatment of multidrug-resistant Plasmodium falciparum malaria with 3-day artesunate-mefloquine combination. *J Infect Dis* 1994;**170**:971–7.
62 **Looareesuwan S**, Vanijanonta S, Viravan C, *et al*. Randomized trial of mefloquine alone and artesunate followed by mefloquine for the treatment of acute uncomplicated falciparum malaria. *Ann Trop Med Parasitol* 1994;**88**:131–6.
63 **Dorsey G**, Vlahos J, Kamya MR, *et al*. Prevention of increasing rates of treatment failure by combining sulfadoxine-pyrimethamine with artesunate or amodiaquine for the sequential treatment of malaria. *J Infect Dis* 2003;**188**:1231–8.
64 **Sutherland CJ**, Drakeley CJ, Obisike U, *et al*. The addition of artesunate to chloroquine for treatment of Plasmodium falciparum malaria in Gambian children delays, but does not prevent treatment failure. *Am J Trop Med Hyg* 2003;**69**:19–25.
65 **Adjuik M**, Babiker A, Garner P, *et al*. Artesunate combinations for treatment of malaria: meta-analysis. *Lancet* 2004;**363**:9–17.
66 **Olliaro PL**, Taylor WR. Developing artemisinin based drug combinations for the treatment of drug resistant falciparum malaria: a review. *J Postgrad Med* 2004;**50**:40–4.
67 **Newton P**, Proux S, Green M, *et al*. Fake artesunate in southeast Asia. *Lancet* 2001;**357**:1948–50.
68 **van Vugt M**, Thaiaporn I, Chanthapadith K, *et al*. Artemether-lumefantrine for the treatment of multidrug-resistant falciparum malaria. *Trans R Soc Trop Med Hyg* 2000;**94**:545–8.
69 **Angus BJ**, Thaiaporn I, Chanthapadith K, *et al*. Oral artesunate dose-response relationship in acute falciparum malaria. *Antimicrob Agents Chemother* 2002;**46**:778–82.
70 **Newton CR**, Krishna S. Severe falciparum malaria in children: current understanding of pathophysiology and supportive treatment. *Pharmacol Ther* 1998;**79**:1–53.
71 **Pukrittayakamee S**, Supanaranond W, Looareesuwan S, *et al*. Quinine in severe falciparum malaria: evidence of declining efficacy in Thailand. *Trans R Soc Trop Med Hyg* 1994;**88**:324–7.
72 **Tran TH**, Day NP, Nguyen HP, *et al*. A controlled trial of artemether or quinine in Vietnamese adults with severe falciparum malaria. *N Engl J Med* 1996;**335**:76–83.
73 **Karbwang J**, Tin T, Rimchala W, *et al*. Comparison of artemether and quinine in the treatment of severe falciparum malaria in south-east Thailand. *Trans R Soc Trop Med Hyg* 1995;**89**:668–71.
74 **van Hensbroek MB**, Onyiorah E, Jaffar S, *et al*. A trial of artemether or quinine in children with cerebral malaria. *N Engl J Med* 1996;**335**:69–75.
75 **Murphy S**, English M, Waruiru C, *et al*. An open randomized trial of artemether versus quinine in the treatment of cerebral malaria in African children. *Trans R Soc Trop Med Hyg* 1996;**90**:298–301.
76 **Walker O**, Salako LA, Omokhodion SI, *et al*. An open randomized comparative study of intramuscular artemether and intravenous quinine in cerebral malaria in children. *Trans R Soc Trop Med Hyg* 1993;**87**:564–6.
77 **Danis M**, Chandenier J, Doumbo O. Results obtained with i.m. artemether versus i.v. quinine in the treatment of severe malaria in a multicentre study in Africa. *Jpn J Trop Med Hyg* 1996;**24**:93–6.
78 **Taylor TE**, Wills BA, Kazembe P, *et al*. Rapid coma resolution with artemether in Malawian children with cerebral malaria. *Lancet* 1993;**341**:661–2.
79 **Pittler MH**, Ernst E. Artemether for severe malaria: a meta-analysis of randomized clinical trials. *Clin Infect Dis* 1999;**28**:597–601.
80 **McIntosh HM**, Olliaro P. Artemisinin derivatives for treating severe malaria. *Cochrane Library*. Issue 2. Oxford: Update Software, 2000.
81 **Artemether-Quinine Meta-analysis Study Group**. A meta-analysis using individual patient data of trials comparing artemether with quinine in the treatment of severe falciparum malaria. *Trans R Soc Trop Med Hyg* 2001;**95**:637–50.
82 **Hien TT**, Phu NH, Mai NT, *et al*. An open randomized comparison of intravenous and intramuscular artesunate in severe falciparum malaria. *Trans R Soc Trop Med Hyg* 1992;**86**:584–5.
83 **Cao XT**, Bethell DB, Pham TP, eet al. Comparison of artemisinin suppositories, intramuscular artesunate and intravenous quinine for the treatment of severe childhood malaria. *Trans R Soc Trop Med Hyg* 1997;**91**:335–42.
84 **Ha V**, Nguyen NH, Tran TB, *et al*. Severe and complicated malaria treated with artemisinin, artesunate or artemether in Viet Nam. *Trans R Soc Trop Med Hyg* 1997;**91**:465–7.
85 **Agbenyega T**, Angus BJ, Bedu-Addo G, *et al*. Glucose and lactate kinetics in children with severe malaria. *J Clin Endocrinol Metab* 2000;**85**:1569–76.
86 **Barennes H**, Sterlingot H, Nagot N, *et al*. Intrarectal pharmacokinetics of two formulations of quinine in children with falciparum malaria. *Eur J Clin Pharmacol* 2003;**58**:649–52.
87 **Arnold K**, Tran TH, Nguyen TC, *et al*. A randomized comparative study of artemisinine (qinghaosu) suppositories and oral quinine in acute falciparum malaria. *Trans R Soc Trop Med Hyg* 1990;**84**:499–502.
88 **Navaratnam V**, Mansor SM, Mordi MN, *et al*. Comparative pharmacokinetic study of oral and rectal formulations of artesunic acid in healthy volunteers. *Eur J Clin Pharmacol* 1998;**54**:411–14.
89 **Barnes KI**, Mwenechanya J, Tembo M, *et al*. Efficacy of rectal artesunate compared with parenteral quinine in initial treatment of moderately severe malaria in African children and adults: a randomised study. *Lancet* 2004;**363**:1598–605.
90 **Karbwang J**, Na-Bangchang K, Wattanakoon Y, *et al*. Artemether 5 versus 7 day regimen for severe falciparum malaria. *Southeast Asian J Trop Med Public Health* 1994;**25**:702–6.
91 **Giao PT**, Binh TQ, Kager PA, *et al*. Artemisinin for treatment of uncomplicated falciparum malaria: is there a place for monotherapy? *Am J Trop Med Hyg* 2001;**65**:690–5.
92 **White NJ**, Nosten F, Looareesuwan S, *et al*. Averting a malaria disaster. *Lancet* 1999;**353**:1965–7.
93 **White NJ**. Antimalarial drug resistance. *J Clin Invest* 2004;**113**:1084–92.
94 **Brewer TG**, Grate SJ, Peggins JO, *et al*. Fatal neurotoxicity of arteether and artemether. *Am J Trop Med Hyg* 1994;**51**:251–9.
95 **Brewer TG**, Peggins JO, Grate SJ, *et al*. Neurotoxicity in animals due to arteether and artemether. *Trans R Soc Trop Med Hyg* 1994;**88**(suppl 1):S33–6.
96 **Van Vugt M**, Angus BJ, Price RN, *et al*. A case-control auditory evaluation of patients treated with artemisinin derivatives for multidrug-resistant Plasmodium falciparum malaria. *Am J Trop Med Hyg* 2000;**62**:65–9.
97 **Kissinger E**, Hien TT, Hung NT, *et al*. Clinical and neurophysiological study of the effects of multiple doses of artemisinin on brain-stem function in Vietnamese patients. *Am J Trop Med Hyg* 2000;**63**:48–55.
98 **Hien TT**, Turner GD, Mai NT, *et al*. Neuropathological assessment of artemether-treated severe malaria. *Lancet* 2003;**362**:295–6.
99 **Gordi T**, Lepist EI. Response to the letter to the editor by Stephen Toovey and Andrew Jamieson. *Toxicol Lett* 2004;**151**:491–2.
100 **Li QG**, Brueckner RP, Peggins JO, *et al*. Arteether toxicokinetics and pharmacokinetics in rats after 25 mg/kg/day single and multiple doses. *Eur J Drug Metab Pharmacokinet* 1999;**24**:213–23.
101 **Teja-Isavadharm P**, Nosten F, Kyle DE, *et al*. Comparative bioavailability of oral, rectal, and intramuscular artemether in healthy subjects: use of simultaneous measurement by high performance liquid chromatography and bioassay. *Br J Clin Pharmacol* 1996;**42**:599–604.
102 **Gordi T**, Lepist EI. Artemisinin derivatives: toxic for laboratory animals, safe for humans? *Toxicol Lett* 2004;**147**:99–107.
103 **Toovey S**, Jamieson A. Audiometric changes associated with the treatment of uncomplicated falciparum malaria with co-artemether. *Trans R Soc Trop Med Hyg* 2004;**98**:261–7, 268–9.
104 **Leonardi E**, Gilvary G, White NJ, *et al*. Severe allergic reactions to oral artesunate: a report of two cases. *Trans R Soc Trop Med Hyg* 2001;**95**:182–3.
105 **Orjih AU**. Haemolysis of Plasmodium falciparum trophozoite-infected eryuthrocytes after artemisinin exposure. *Br J Haematol* 1996;**92**:324–8.
106 **Vennerstrom JL**, Arbe-Barnes S, Brun R, *et al*. Identification of an antimalarial synthetic trioxolane drug development candidate. *Nature* 2004;**430**:900–4.
107 **Xiao SH**, Booth M, Tanner M. The prophylactic effects of artemether against Schistosoma japonicum infections. *Parasitol Today* 2000;**16**:122–6.
108 **Utzinger J**, N'Goran EK, N'Dri A, *et al*. Oral artemether for prevention of Schistosoma mansoni infection: randomised controlled trial. *Lancet* 2000;**355**:1320–5.
109 **Efferth T**, Sauerbrey A, Olbrich A, *et al*. Molecular modes of action of artesunate in tumor cell lines. *Mol Pharmacol* 2003;**64**:382–94.
110 **White NJ**, Krishna S. Treatment of malaria: some considerations and limitations of the current methods of assessment. *Trans R Soc Trop Med Hyg* 1989;**83**:767–77.

ANSWERS

1. (A)—false: recrudescence rates are generally 10%–20% even when courses are extended to seven days. (B)—false: millions of doses of artemisinins have been given without any firm evidence of neurotoxicity in humans. (C)—false: this has not been shown in Africa or Asia. (D)—true: their short half life renders them inappropriate for this indication. (E)—false: in this area of multidrug resistance there is no evidence of in vivo resistance despite extensive use for over a decade. (F)—false: de novo synthesis is difficult and artemisinins remain entirely plant derived (from *Artemisia annua* plantations in Asia). 2. Intramuscular and intravenous artesunate (intramuscular administration of artemether and arteether show pharmacokinetic disadvantages in terms of the speed and reliability of absorption). Trials of parenteral artesunate compared with quinine are in progress. Rectal formulation of artesunate also shows promise for use in treating moderate malaria in rural settings.