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Two fixed-dose artemisinin combinations for drug-resistant falciparum and vivax malaria in Papua, Indonesia: an open-label randomised comparison

A Ratcliff, H Siswanto, E Kenangalem, R Maristela, R M Wuwung, F Laihad, E P Ebsworth, N M Anstey, E Tjitra, and R N Price

International Health Programme, Menzies School of Health Research and Charles Darwin University, Darwin, Northern Territory, Australia (A Ratcliff FRACP, Prof N M Anstey FRACP, R N Price FRCP); National Institute of Health Research and Development, Ministry of Health, Jakarta, Indonesia (H Siswanto MD, E Tjitra MD); MSHR-NIHRD Malaria Research Programme, Timika, Indonesia (E Kenangalem MD); District Ministry of Health, Timika, Papua, Indonesia (E Kenangalem); Public Health and Malaria Control Department, PT Freeport, Indonesia, Tembagapura, Papua, Indonesia (R Maristela MD, R M Wuwung, E P Ebsworth MPH); International SOS, Tembagapura, Papua, Indonesia (R Maristela, E P Ebsworth); Directorate General of Centre for Disease Control and Environmental Health, Ministry of Health, Jakarta, Indonesia (F Laihad MD); Centre for Vaccinology and Tropical Medicine, Nuffield Department of Clinical Medicine, John Radcliffe Hospital, Oxford, UK (R N Price)

Summary

Background—The burden of *Plasmodium vivax* infections has been underappreciated, especially in southeast Asia where chloroquine resistant strains have emerged. Our aim was to compare the safety and efficacy of dihydroartemisinin-piperaquine with that of artemether-lumefantrine in patients with uncomplicated malaria caused by multidrug-resistant *P falciparum* and *P vivax*.

Methods—774 patients in southern Papua, Indonesia, with slide-confirmed malaria were randomly assigned to receive either artemether-lumefantrine or dihydroartemisinin-piperaquine and followed up for at least 42 days. The primary endpoint was the overall cumulative risk of parasitological failure at day 42 with a modified intention-to-treat analysis. This trial is registered with ClinicalTrials.gov, trial number 00157833.

Findings—Of the 754 evaluable patients enrolled, 466 had infections with *P falciparum*, 175 with *P vivax*, and 113 with a mixture of both species. The overall risk of failure at day 42 was 43% (95% CI 38–48) for artemether-lumefantrine and 19% (14–23) for dihydroartemisinin-piperaquine (hazard ratio=3.0, 95% CI 2.2–4.1, $p<0.0001$). After correcting for reinfections, the risk of recrudescence of *P falciparum* was 4.4% (2.6–6.2) with no difference between regimens. Recurrence of vivax occurred in 38% (33–44) of patients given artemether-lumefantrine compared

Correspondence to: Dr Ric Price, Menzies School of Health Research, PO Box 41096, Casuarina, Darwin, NT 0811 Australia
rnp@menzies.edu.au.

Contributors

Alison Ratcliff, Hadjar Siswanto, Enny Kenangalem, Maxi Wuwung, and Rilia Maristela all participated in undertaking the clinical studies. The trial was coordinated and supervised by Emiliana Tjitra, Peter Ebsworth, Nick Anstey, and Ric Price. Ferdinand Laihad supervised the research programme. All authors participated in the design, analysis, interpretation, and writing up of the research work.

Conflict of interest statement

RP has received reimbursement of travel expenses to attend meetings by Novartis, who manufacture Coartem, and is on a data safety monitoring committee for Sygma Tau, who manufacture dihydroartemisinin-piperaquine.

with 10% (6.9–14.0) given dihydroartemisinin-piperaquine ($p < 0.0001$). At the end of the study, patients receiving dihydroartemisinin-piperaquine were 2.0 times (1.2–3.6) less likely to be anaemic and 6.6 times (2.8–16) less likely to carry vivax gametocytes than were those given artemether-lumefantrine.

Interpretation—Both dihydroartemisinin-piperaquine and artemether-lumefantrine were safe and effective for the treatment of multidrug-resistant uncomplicated malaria. However, dihydroartemisinin-piperaquine provided greater post-treatment prophylaxis than did artemether-lumefantrine, reducing *P falciparum* reinfections and *P vivax* recurrences, the clinical public-health importance of which should not be ignored.

Introduction

Resistance to antimalarial drugs is emerging throughout tropical regions. WHO recommends the use of artemisinin combination therapies to improve antimalarial effectiveness and keep the selection of drug-resistant parasites to a minimum. Although more than 60 countries now advocate changing antimalarial policy to artemisinin combination therapies, debate still continues as to the most suitable combination and how these new treatments should be deployed and funded.

Southeast Asia has been a focus for emergence of drug resistant strains of *Plasmodium falciparum* for more than 40 years, but drug resistance in *Plasmodium vivax* has evolved slower than in *P falciparum*. The first chloroquine resistant isolates of *P vivax* were reported from Papua, Indonesia, and Papua New Guinea,^{1,2} and cases have also been reported from Myanmar, India and South America.^{3–5} In a study from southern Papua, the risk of failure within 28 days was 65% after chloroquine monotherapy for *P vivax* and 48% after chloroquine plus sulfadoxine and pyrimethamine for *P falciparum*.⁶ These risks are similar to those in studies from north Papua.^{7,8} Virtually no published data exist as to how best to treat chloroquine resistant *P vivax*.^{9,10} Since most patients in endemic countries are diagnosed and treated without differentiation of the species of infection and since the risk of early relapse from the hypnozoite liver stages after treatment of *P vivax* or even *P falciparum* (alone) infections often exceeds 40%,¹¹ it is becoming apparent that we need to quantify the burden of recurrent *P vivax* infection and investigate alternative treatment strategies.

Artemether-lumefantrine is the only widely available coformulated artemisinin combination therapy that is produced to international good manufacturing practice standards, and has therefore become the preferred choice in many endemic countries. The only other widely available fixed-dosed treatment is dihydroartemisinin-piperaquine. The antimalarial activity of piperaquine has been recognised since the 1960s, although its use in clinical practice has been restricted mainly to China, where piperaquine replaced chloroquine in the national malaria control programme in 1978. In the past 5 years there has been evidence that dihydroartemisinin-piperaquine is effective against multidrug-resistant strains of *P falciparum*.^{12–15}

In Papua, where drug resistance has emerged to both *P falciparum* and *P vivax*, we undertook a randomised comparative study to establish the safety and efficacy of artemether-lumefantrine and dihydroartemisinin-piperaquine for the treatment of patients with uncomplicated malaria, who presented to rural clinics with infection with *P falciparum*, *P vivax*, or a mixture of both species.

Methods

Study site and design

Our study was undertaken in two established rural outpatient clinics, 6 km apart and 20 km west of the city of Timika, in south Papua, Indonesia. This forested lowland area has unstable malaria transmission associated with three mosquito vectors: *Anopheles koliensis*, *A farauti*, and *A Punctulatus*.^{16,17} The yearly incidence of malaria in the region is 938 per 1000 person-years, with a *P falciparum* and *P vivax* infection ratio of 57/43. Both species have a high rate of chloroquine resistance.⁶ Ethnic origin of the local population is diverse because of economic migration, with highland Papuans, lowland Papuans, and non-Papuans all living in the region. In view of the high number of infections in non-immune patients, local protocols recommend that all patients with patent parasitaemia are given antimalarial therapy. Previous studies have reported that the pyrogenic density in this region is 1000/ μ L for *P falciparum* and 400/ μ L for *P vivax*.^{7,9}

We did a prospective open-label randomised comparison of artemether-lumefantrine (Coartem, Novartis Pharma, Basel, Switzerland) with dihydroartemisinin-piperaquine (Artekin, Holleykin Pharmaceuticals, Guangzhou, China) for the treatment of infections with *P falciparum*, *P vivax*, or a mixture of both species, in children and adults with uncomplicated symptomatic malaria. Our study was based on the 2003 WHO in-vivo antimalarial drug sensitivity protocol,¹⁸ which was modified to include mixed infections and any parasitaemia. Patients were followed up for 42 days with a standard clinical record form for drug effectiveness.

The primary endpoint was the overall cumulative risk of any parasitaemia reappearing during the 42-day follow-up. Previous studies have shown that 95% of true recrudescences after artemisinin combination therapies will take place within this time.¹⁹ Secondary endpoints included the risk of reappearance of *P falciparum*, *P vivax*, and true recrudescence *P falciparum*, and the proportion of parasitaemic patients on days 1, 2, and 3, post-treatment gametocyte carriage, and haematological recovery. When parasite count, gametocyte carriage, or haemoglobin were missing, numbers were restricted to patients with acceptable data presenting on specific days. Although this study was open labelled, the final endpoints were determined by microscopic confirmation and PCR analysis, which were done blind to treatment allocation.

The study was approved by the Ethics committee of the National Institute of Health Research and Development, Indonesian Ministry of Health (Jakarta, Indonesia), the Ethics committee of Menzies School of Health Research (Darwin, Australia), and the Oxford Tropical Research Ethics Committee (Oxford, UK). Written informed consent was obtained from adult patients and parents of enrolled children. The trial was registered with clinicaltrials.gov, number NCT 00157833.

Patients

Patients with slide-confirmed malaria (*P falciparum*, *P vivax*, or mixed infections) and fever or a history of fever during the preceding 48 h, who presented to the outpatient clinic, were eligible for enrolment. Pregnant or lactating women and children under 10 kg were excluded, as were patients with WHO danger signs or signs of severity,²⁰ a parasitaemia greater than 4%, or concomitant disease requiring hospital admission. To focus resources and ensure adequate follow-up of the patients enrolled, we restricted recruitment to a maximum of five patients per day from each clinic.

Procedures

After enrolment, patients were randomly assigned to receive either artemether-lumefantrine or dihydroartemisinin-piperaquine. A randomisation list was generated in blocks of 20 patients by an independent statistician, with each treatment allocation concealed in an opaque sealed envelope that was opened once the patient had been enrolled. A standard data sheet was completed to record demographic information, details of symptoms and their duration, and previous antimalarial medication. Clinical examination findings were documented, including the axillary temperature that was measured with a digital thermometer. Venous blood was taken for blood film, haematocrit (packed cell volume), and white cell count. Parasite counts were done on Giemsa-stained thick films as the number of parasites per 200 white blood cells, and peripheral parasitaemia was calculated on the assumption of a white cell count of 7300/ μ L. All slides were read by a certified microscopist with at least 10 years experience, who was blinded to treatment allocation. A thick smear was regarded as negative on initial review if no parasites were seen in 100 high power fields. A thin smear was also examined to confirm parasite species and used for quantification if parasitaemia was greater than 200 per 200 white blood cells. To cross check, 200 high power fields were examined before slides were regarded as negative. All slides were cross checked by a second experienced microscopist. In 9% of cases, in which readings were discordant with the first reading, slides were reread by a third microscopist and a consensus reached.

Patients were examined every day thereafter until they became afebrile and parasite free. A blood smear was taken and a symptom questionnaire completed at every visit. Patients were then seen every week for 6 weeks. At each clinic appointment a full physical examination was done, the symptom questionnaire completed, and blood taken to check for parasite count and haemoglobin concentration with a battery operated portable photometer (HemoCue Hb201+, Angelholm, Sweden). Blood spots on filter paper (Whatman chromatography paper [Whatman BFC 180, Maidstone, UK]) were also obtained at enrolment and day of failure.

Artemether-lumefantrine was dispensed according to patients' weight. Patients weighing 10–15 kg received one tablet per dose, those weighing 15–25 kg received two, those of 25–35 kg received three, and those greater than 35 kg received four. In total, six doses were given over 3 days: on admission and at 8, 24, 36, 48, and 60 h. The morning dose was supervised and the patient then given the evening dose to take themselves.

Dihydroartemisinin-piperaquine was given as a weight per dose regimen of 2.25 mg/kg and 18 mg/kg per dose of dihydroartemisinin and piperaquine, respectively, rounded up to the nearest half tablet. All doses were supervised and given on admission, after 24 h, and at 48 h.

Since the bioavailability of both lumefantrine and piperaquine is increased if taken with a fatty meal,^{21,22} patients were instructed to take every dose with a biscuit or milk. When drug administration was observed and vomiting happened within 60 minutes, the full dose was given again. If the axillary temperature was greater than 38°C, paracetamol was given. Patients failing therapy with recurrence of *P. falciparum* were retreated with quinine (10 mg of salt per kg of body weight, given orally three times a day for 7 days) with additional doxycycline (100 mg twice a day for 7 days) if the patient was older than 8 years. Patients with reappearance of *P. vivax* were offered a 3-day course of supervised amodiaquine (Flavoquine, Aventis, Paris, France. 153 mg base per tablet, 30 mg/kg over 3 days) or, if they refused to return for supervised therapy, quinine with or without doxycycline. Indonesian protocols recommend the use of primaquine (0.3 mg of base/kg of bodyweight

for 14 days) for patients with *P vivax* infection, but to decrease potential drug interaction, administration was delayed until day 28 of the study.

Statistical analysis

Statistical analysis was done accordingly to an a-priori analytical plan, in which estimates of efficacy were derived from all patients who did not violate any of the inclusion or exclusion criteria (ie, modified intention-to-treat).

In a pilot study, 60% of patients presenting to the clinics with uncomplicated malaria had *P falciparum*, 25% had *P vivax*, and 15% had mixed infections. On the assumption that 96% of patients are cured after artemether-lumefantrine or dihydroartemisinin-piperaquine is given fully supervised, a total sample size of 750 patients would record the risk of failure for each regimen within 3% that of the true risk (allowing for 30% loss during follow-up). Furthermore, the study had 80% power and 95% confidence to detect a 10% difference in the risk of failure that might have resulted from a policy in which drug administration was supervised only once daily (a practical constraint of widespread deployment).

Data were double entered and validated with EpiData software (version 3.02) and analysed with SPSS for Windows (version 14). The Mann-Whitney U test or Kruskal-Wallis method were used for non-parametric comparisons, and *t* test or one-way analysis of variance for parametric comparisons. For categorical variables, percentages and 95% CIs were calculated by Wilson's method. Proportions were examined by χ^2 test with Yates' correction or by Fisher's exact test.

The cumulative risk of failure was assessed by survival analysis with the Kaplan Meier method on a modified intention to treat basis. Anyone lost during follow-up or, in the case of secondary endpoints, presenting with a different outcome, were censored on their last day of follow-up and regarded as not being treatment failures. Patients failing to complete a 42-day follow-up for any other reason were regarded as treatment failures. Those with recurrent vomiting or adverse drug effects, who required early cessation of treatment and rescue therapy, were regarded as early therapeutic failures. Groups were compared by use of the Mantel-Haenszel log rank test and the hazard ratio (HR) was presented. Furthermore, treatment outcomes were compared and HR calculated after stratification for the initial infecting parasite species with Cox proportional hazards model.

In patients with *P falciparum* (alone or mixed infection) in both initial and recurrent parasitaemia, polymorphisms within the genes that encode MSP-1 (merozoite specific protein), MSP-2, and GLURP (glutamate-rich protein) were used to record reinfections and recrudescence infections, with PCR methods described previously.²³ In total, 43 patients had paired infections with *P falciparum* initially (alone or mixed) and at the time of recurrence. Of the 70% (30/43) of these paired samples that could be interpreted, 87% (26/30) were reinfections. Hence, in the 30% (13/43) of cases in whom the PCR result was unavailable or uninterpretable the reappearance of *P falciparum* was regarded, on balance, as reinfection. When the risk of recrudescence *P falciparum* was calculated, all early treatment failures and true recrudescences were regarded as failures, but *P falciparum* reinfections and recurrences of *P vivax* were censored as non-failures in survival analysis.

Gametocyte carriage was assessed by calculation of person gametocyte week rates as a measure of transmission potential. These rates were defined as the number of weeks in which blood slides were positive for gametocytes during 6-week follow-up, divided by the total number of weeks followed up, and were expressed per 1000 person-weeks.²⁴ Adverse events reported were generally associated with acute malaria, and hence comparison

between treatment groups was made in patients without the symptom on admission but who developed the complaint after starting antimalarial treatment.

Role of the funding source

The sponsors of the study had no role in study design, data collection, data analysis, data interpretation, or writing of the report. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit for publication.

Results

Between July, 2004, and June, 2005, 3473 patients with uncomplicated malaria were treated at the recruitment clinics. For the 2992 (86%) patients meeting the eligibility criteria, age and sex distribution did not differ between the 774 (26%) patients enrolled and the whole clinic workload. There were 20 protocol violations all of which were identified within 24 h, offered alternative treatment, and excluded from further analysis (figure 1). Of the remaining 754 patients, 466 (62%) had only *P falciparum* infection, 175 (23%) had *P vivax* infection, and 113 (15%) had both species present. Table 1 shows the baseline characteristics. Overall, follow-up to day 42 or day of failure was achieved in 78% (293/375) of patients given artemether-lumefantrine and 72% (274/379) given dihydroartemisinin-piperaquine ($p=0.076$).

Seven (1%) of the 754 patients correctly enrolled were unable to tolerate their drug because of recurrent vomiting (figure 1). Two patients developed urticaria on day 1, and one developed a non-specific erythematous rash on day 2. These patients were regarded as treatment failures and were given quinine either orally or intravenously. They all had unremarkable recoveries.

Early clinical deterioration that needed hospital treatment was seen in two patients. A 3-year-old boy with only *P falciparum* infection developed respiratory distress, prostration, and profuse diarrhoea 24 h after his first dose of artemether-lumefantrine. He was admitted, was given intravenous fluids and quinine, and made a full recovery. A 5-year-old girl with mixed parasitaemia and high fever vomited after her first dose of dihydroartemisinin-piperaquine and 30 minutes later developed a complex partial seizure. She was transferred to hospital where she was given intravenous quinine and made a full recovery.

73% (254/350) of the remaining patients given dihydroartemisinin-piperaquine had cleared their parasites within 24 h, compared with 57% (192/339) of those who received artemether-lumefantrine ($p<0.0001$). By day 2, 97% (690/712) of patients were parasite free and 99% (662/671) were afebrile, with no difference between treatment groups.

A total of 163 patients had recurrent parasitaemia during follow-up, of which 34 infections were attributable to *P falciparum* alone, 102 to *P vivax* alone, two to *P ovale* alone, and 25 contained both species. Overall 53% (85/161) of recurrences were symptomatic, 12% (19/159) had documented fever, and 33% (43/131) were anaemic (haemoglobin <100 g/L). These proportions did not differ between treatment groups or infecting species.

The cumulative risk of any parasitological failure was greater after artemether-lumefantrine than after dihydroartemisinin-piperaquine (HR 3.0, 95% CI 2.2–4.1, $p<0.0001$, table 2). Reappearance of *P falciparum* (alone or mixed) was much the same in the two groups (32 patients after artemether-lumefantrine and 27 after dihydroartemisinin-piperaquine, table 2). Although by day 42 reappearance of *P falciparum* did not differ between treatment groups, the time to reappearance was longer in patients receiving dihydroartemisinin-piperaquine (median 38 days, range 21–45) than in those receiving artemether-lumefantrine (34 days,

19–43), $p=0.018$ (figure 2). The risk of recurrent *P falciparum* infections was not dependent on the species of the initial infection. After PCR correction, the risk of true recrudescent *P falciparum* during follow-up was less than 5% in both treatment groups (table 2).

Recurrence of *P vivax* (alone or mixed) was recorded in 127 patients. By day 42, the cumulative risk of *P vivax* was greater in patients given artemether-lumefantrine than in those given dihydroartemisinin-piperaquine (38% vs 10%, table 2, figure 2). The cumulative risk of *P vivax* recurrence took place in 36% (95%CI 28–44) of patients initially infected with *P vivax*, 17% (13–21) of those with *P falciparum*, and 36% (26–46) of those with mixed infections ($p<0.0001$). After stratification by the species at presentation, the hazard ratio for *P vivax* reappearance associated with artemether-lumefantrine was 4.8 (95%CI 3.2–7.2), $p<0.0001$ (table 2).

On admission, *P falciparum* gametocytes were present in 21% (122/574) of patients with *P falciparum* alone or mixed infections. In patients without gametocytes on presentation, the falciparum gametocyte carriage during follow-up was 5.7 per 1000 person-weeks, with no difference between treatment groups.

P vivax gametocytes on admission were present in 56% (160/284) of patients with *P vivax* alone or mixed infections. After day 7, *P vivax* gametocytaemia was always associated with recurrence of the organism's asexual stages, with carriage occurring at a rate of 3.7 per 1000 patient-weeks after dihydroartemisinin-piperaquine compared with 24.6 after artemether-lumefantrine (rate ratio [RR]=6.6, 95%CI 2.8–16.0, $p=0.0002$) (figure 3).

Baseline haemoglobin concentrations were assessed in 99% (750/754) of patients, 387 (52%) of whom had anaemia (haemoglobin <100 g/L). Haemoglobin concentrations were available in 42% (1403/3326) of the subsequent reviews of patients, with no difference between treatment groups. Although the mean haemoglobin concentrations initially rose after treatment (figure 4), by the end of the study the mean haemoglobin in patients with a recurrent parasitaemia was 10.2 g/L (95% CI 4.4–16.0) lower than those successfully treated ($p<0.0001$). The corresponding risk of anaemia was 33% (45/135) in patients failing therapy compared with 19% (21/111) in those successfully treated (RR=1.8, 95% CI 1.1–2.8, $p=0.02$). By day 42, 35% (24/68) of patients given artemether-lumefantrine were anaemic, compared with 17% (15/86) of those who received dihydroartemisinin-piperaquine (relative risk 2.0, 1.2–3.6, $p=0.019$).

Early vomiting within the first hour of drug administration was recorded in 2.7% (ten of 375) of patients given artemether-lumefantrine and 2.9% (11/379) of patients given dihydroartemisinin-piperaquine. 11% (11/96) of children less than 5 years of age vomited the drug, compared with 3% (seven of 227) in older children, and 1% (three of 431) in adults ($p<0.0001$).

Adverse events were assessed in patients without the symptom at enrolment (figure 5). The only difference between the two treatment groups was a two-fold (95% CI 1.3–3.3) increased risk of diarrhoea on days 1 and 2, which arose in more patients receiving dihydroartemisinin-piperaquine than in those given artemether-lumefantrine. By day 7, the risk of diarrhoea was 5% in both treatment groups. Although 35% (12/34) of patients developed a headache on days 1 and 2 after dihydroartemisinin-piperaquine (compared with 23% [nine of 40] of patients given artemether-lumefantrine), the difference was not significant since headache was a common symptom at presentation.

The three patients who developed urticaria (two on day 1 and one on day 2 [after his final dose and not included as treatment failure]) were treated with antihistamines and all made a complete recovery. A 3-year-old child who developed an erythematous rash on day 1 was

treated with antihistamines and the rash had resolved by day 4. A 32-year-old man died suddenly and unexpectedly 60 days after treatment with artemether-lumefantrine. He had been assessed on day 28 and was reported as being well—analysis of his blood on day 28 revealed no haematological or biochemical abnormalities. No further details were available.

Of the 163 patients with recurrent parasitaemia, 139 (85%) agreed to be retreated and followed up for a further 28 days. Of the 48 patients representing with *P falciparum* (alone or mixed infections), who were given an unsupervised course of quinine with or without doxycycline, the risk of failure at day 28 rose to 83% (95% CI 70–96). Of the 91 patients with a recurrence of *P vivax* alone, 26 patients were retreated with an unsupervised course of quinine plus primaquine with or without doxycycline, with a subsequent risk of failure of 57% (36–79). The remaining 65 patients with *P vivax* were retreated with a supervised course of amodiaquine plus primaquine. Two patients had recurrent vomiting, four failed to complete therapy, and 18 had recurrent parasitaemia by day 28 (one with *P falciparum*, nine with *P vivax*, and eight with mixed infections). By day 28, the cumulative risk of recurrence with *P vivax* was 26% (12–40).

Discussion

Our pragmatic study design is novel, enrolling and randomising patients infected with either species and then quantifying the subsequent risk of recurrence and associated morbidity. We have assessed the efficacy of two widely available fixed-dose artemisinin combinations for patients with drug-resistant *P falciparum* and *P vivax*. Both artemether-lumefantrine and dihydroartemisinin-piperaquine were well tolerated and associated with a rapid clinical response. Although the initial parasite clearance was significantly faster after dihydroartemisinin-piperaquine, almost all patients in both groups were parasite free and afebrile within 48 h. However, by the end of follow-up, the cumulative risk of parasitological failure was significantly higher after artemether-lumefantrine than after dihydroartemisinin-piperaquine. Over half these recurrent parasitaemias were associated with clinical symptoms and a third with anaemia, with *P falciparum* and *P vivax* malaria associated with equal morbidity.

The risk of true recrudescence with *P falciparum* was less than 5% in both treatment groups, and most *P falciparum* recurrences were caused by reinfections. These findings are in keeping with previous efficacy studies of artemether-lumefantrine^{25,26} and dihydroartemisinin-piperaquine^{12–15} and suggest that most patients, even those who were only partly supervised, completed a full course of treatment.

Overall, most recurrent infections were attributable to *P vivax*, which could have arisen from recrudescence from the same isolate, reinfection with a new isolate, or relapse from hypnozoite stages.²⁷ Although there are as yet no reliable genetic methods for discerning between these possibilities, the discrepancy between the cumulative risk of failure probably arose as a result of the difference between the terminal elimination half-life of lumefantrine (around 4 days)²¹ and that of piperaquine (28–35 days).^{28,29} Artemisinin combinations achieve their antimalarial effect through an initial rapid reduction in parasite biomass attributable to the short-term but potent artemisinin derivative, with the subsequent removal of the remaining parasites by the intrinsically less active but more slowly eliminated lumefantrine or piperaquine. After eradication of the asexual stages of the parasite from peripheral blood, patients who remain in an endemic area are at risk of reinfection or, in *P vivax* endemic areas, from relapses from the liver stage hypnozoites. Slowly eliminated antimalarial drugs will exert a greater post-treatment prophylactic effect than those that are more rapidly eliminated, and this enhanced effect is seen in both the rates of reinfection and

relapse. The greater the risk of *P. vivax* relapse or reinfection with either species, the more apparent this prophylactic effect will probably become.

In our study, the first *P. vivax* recurrence took place 17 days after artemether-lumefantrine was given. By contrast, the first recurrence after dihydroartemisinin-piperaquine did not take place until day 29. The delay in *P. falciparum* reinfection was also marked, but smaller than for *P. vivax*. We were unable to follow up patients for any longer than 6 weeks, but if we had done so the survival curves would probably have merged over time as the plasma concentrations of piperaquine fell. However, the delay in relapse and reinfections conferred by dihydroartemisinin-piperaquine gave patients a lengthened period without symptomatic malaria, allowing for an increased time for haematological recovery, which in turn halved the risk of anaemia. The reduction in the recurrence of *P. vivax* infections also substantially reduced the transmission potential to the mosquito vector. Although the public-health implications of these results need to be confirmed by longer follow-up and with repeated exposure, our findings suggest that further benefits will accrue with repeated treatments, and that the benefit will be more apparent in patients at greatest risk of infection.

The major concern with deployment of antimalarial drugs with long half-life and combinations with pharmacokinetic mismatching is that there will be an increased risk of selecting drug-resistant isolates.³⁰ Indeed, the long subtherapeutic tail of the antifolate sulfadoxine-pyrimethamine has been directly implicated in the rapid spread of resistance to this agent throughout Asia and Africa.³¹ The emergence and spread of resistant parasites is determined by two major components: selection de novo and selective transmission. Emergence of resistant parasites has been argued to be a function of biomass and therefore will probably take place in the initial infection rather than the reinfecting strains.³² In vivo, the rapid reduction of an infecting biomass with an artemisinin derivative reduces the number of asexual parasites exposed to the second drug, which thus reduces the chances of a resistant mutant emerging during treatment.³²

If dihydroartemisinin-piperaquine is deployed in an area before the emergence of piperaquine resistance, and if the artemisinin component can substantially delay the emergence of de-novo resistance, then the tangible benefits accrued by the longacting combination favour dihydroartemisinin-piperaquine. Once resistance strains emerge in an area, selective transmission in the long subtherapeutic tail of this drug is likely to increase the spread of resistance and herald the demise of the regimen. Careful monitoring of in-vivo and in-vitro antimalarial effectiveness should therefore remain a priority.

Neither artemether-lumefantrine nor dihydroartemisinin-piperaquine affects the hypnozoite stages of *P. vivax*. Early administration of primaquine, the only registered antimalarial agent with activity against hypnozoites, could possibly have prevented the high rates of relapse due to *P. vivax* that we recorded. However, radical cure of *P. vivax* in this area needs high doses of primaquine daily, over 14 days,^{33,34} and in practice such long courses of unsupervised therapy are rarely adhered to. Hence, although the post-treatment prophylaxis afforded by dihydroartemisinin-piperaquine will not prevent subsequent relapses, this drug will provide the only practical means available for delaying these relapses in such areas. Alternative strategies to deal with the hypnozoite stages are needed as a research priority, but as yet are not forthcoming.

In conclusion, artemether-lumefantrine and dihydroartemisinin-piperaquine proved well tolerated, safe, and effective treatments for drug-resistant *P. falciparum*. In Papua, Indonesia, alternative treatment strategies are scarce, but the simple 3-day regimen of dihydroartemisinin-piperaquine, its low price, fast clinical response, and post-treatment prophylactic effect offer substantial benefits over artemether-lumefantrine, the only other

available fixed-dosed artemisinin combination. Although priority should be given to keep the risk of recrudescence to a minimum, the clinical and public health relevance of reinfections and relapses should not be ignored.

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Figure 1. Trial Profile

*Maximum of 5 patients enrolled per clinic each day. †WHO criteria or recurrent vomiting or adverse event warranting rescue therapy.

Figure 2. Cumulative risk of patients failing with *P. falciparum* (upper) and *P. vivax* (lower) parasitaemia (alone or mixed)

Overall difference between treatment groups at day 42, after stratifying for the initial species of infection, for *P. falciparum*, $p=0.186$; for *P. vivax*, $p<0.0001$.

Figure 3.
P. vivax gametocyte carriage in all patients studied after treatment with artemether-lumefantrine or dihydroartemisinin-piperaquine.

Figure 4. Haematological recovery after treatment for patients receiving dihydroartemisinin-piperaquine and artemether-lumefantrine
Error bars=95% CIs.

Figure 5. Adverse events on day 1 or 2 in patients without symptoms on admission
 *p=0.003.

Table 1

Patient characteristics at baseline

	Artemether-lumefantrine (n=375)	Dihydroartemisinin- piperaquine (n=379)
Species at enrolment		
<i>P. falciparum</i> (%)	234 (62%)	232 (61%)
Geometric mean parasitaemia per μL (95%CI)	3790 (2995–4796)	3386 (2674–4289)
Parasitaemia >1000/ μL	177 (76%)	180 (78%)
<i>P. vivax</i>	85 (23%)	90 (24%)
Geometric mean parasitaemia per μL (95%CI)	2143 (1532–2997)	2448 (1729–3465)
Parasitaemia >400/ μL	71 (84%)	74 (82%)
Mixed Infections (%)	56 (15%)	57 (15%)
Geometric mean parasitaemia per μL (95%CI)	4459 (2779–7158)	4304 (2887–6416)
Parasitaemia >400/ μL	50 (89%)	54 (95%)
Male patients	231 (60%)	244 (58%)
Weight (kg) (median, range)	46 (10–104)	48 (10–83)
Age (years) (median, range)	18 (1–60)	17 (1–60)
<5 years	48 (13%)	48 (13%)
5–14 years	109 (29%)	118 (31%)
>14 years	218 (58%)	213 (56%)
Temperature >37.5°C	128 (34%)	118 (31%)
History of malaria in previous month	64 (17%)	74 (20%)
Haemoglobin (g/L) (mean, SD)	98 (25)	98 (24)
Splenomegaly	261 (70%)	248 (65%)

Data are number (%), unless otherwise indicated.

Table 2

Cumulative risk of failure at day 42

	Artemether-lumefantrine	Dihydroartemisinin-piperaquine	Hazard ratio	p value
Parasitological failure with any species				
Initial infection with any species [*]	375 43% (38–48)	379 19% (14–23)	3.0 (2.2–4.1) [‡]	<0.0001
Parasitological failure with <i>P. falciparum</i>				
Initial infection with any species [‡]	375 15% (11–20)	379 13% (9–17)	1.3 (0.9–2.1) [‡]	0.186
True recrudescence of <i>P. falciparum</i> ^{‡,§}	290 4.7% (2.0–7.4)	289 4.1% (CI 1.6–6.6)	1.2 (0.5–2.9)	0.604
Parasitological failure with <i>P. vivax</i>				
Initial infection with any species [‡]	375 38% (33–44)	379 10% (6.9–14.0)	4.8 (3.2–7.2) [‡]	<0.0001
Initial infection with <i>P. vivax</i> (pure or mixed)	141 57% (48–66)	147 14% (7.3–20.0)	5.5 (3.2–9.5)	<0.0001

Efficacy rates were calculated from the modified intent-to-treat population, in which patients failing to complete a 42-day follow-up, other than those lost to follow-up, were regarded as treatment failures (seven in artemether-lumefantrine group and six in dihydroartemisinin-piperaquine group); hence these are conservative estimates for comparative analysis.

Data are n, % (95% CIs).

^{*} Primary endpoint.

[‡] The hazard ratio was calculated with a Cox proportional hazards model after stratifying for the initial species of infection.

[‡] Secondary endpoint.

[§] After correcting by PCR genotyping, patients with *P. falciparum* (alone or mixed) initially and at failure.