

# Combination Therapy for Uncomplicated *Falciparum* Malaria in Ugandan Children

## A Randomized Trial

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**M**ALARIA IS ONE OF THE most important infectious diseases in the world and a leading cause of death in children in Africa.<sup>1</sup> The control of malaria has been challenged by increasing resistance of *Plasmodium falciparum* to antimalarial drugs, particularly chloroquine and sulfadoxine-pyrimethamine, leading to sweeping changes in antimalarial treatment recommendations.<sup>2</sup> Combination regimens, including a number of artemisinin-based combination therapies (ACTs), have replaced monotherapies as the recommended treatments for uncomplicated malaria.<sup>3</sup> However, which regimens offer optimal therapies for malaria in Africa remains unclear.

The World Health Organization (WHO) recommends 5 regimens for the treatment of falciparum malaria, including 4 ACTs and 1 non-ACT regimen.<sup>4</sup> One of the ACT regimens, artesunate + mefloquine, is limited for use because of the expense and poor tolerability of mefloquine. Another, artesunate + sulfadoxine-pyrimethamine, was found to have surprisingly poor efficacy in Uganda and elsewhere.<sup>5,6</sup> The

**Context** Combination therapy is now widely advocated as first-line treatment for uncomplicated malaria in Africa. However, it is not clear which treatment regimens are optimal or how to best assess comparative efficacies in highly endemic areas.

**Objective** To compare the efficacy and safety of 3 leading combination therapies for the treatment of uncomplicated malaria.

**Design, Setting, and Participants** Single-blind randomized clinical trial, conducted between November 2004 and June 2006, of treatment for all episodes of uncomplicated malaria in children in an urban community in Kampala, Uganda. A total of 601 healthy children (aged 1-10 years) were randomly selected and were followed up for 13 to 19 months, receiving all medical care at the study clinic.

**Interventions** Study participants were randomized to receive 1 of 3 combination therapies (amodiaquine plus sulfadoxine-pyrimethamine, amodiaquine plus artesunate, or artemether-lumefantrine) when diagnosed with their first episode of uncomplicated malaria. The same assigned treatment was given for all subsequent episodes.

**Main Outcome Measure** 28-Day risk of parasitological failure (unadjusted and adjusted by genotyping to distinguish recrudescence from new infection) for each episode of uncomplicated malaria treated with study drugs.

**Results** Of enrolled children, 329 of 601 were diagnosed with at least 1 episode of uncomplicated malaria, and 687 episodes of *Plasmodium falciparum* malaria were treated with study drugs. The 28-day risk of treatment failure (unadjusted by genotyping) for individual episodes of malaria were 26.1% (95% CI, 21.1%-32.1%) for amodiaquine plus sulfadoxine-pyrimethamine, 17.4% (95% CI, 13.1%-23.1%) for amodiaquine plus artesunate, and 6.7% (95% CI, 3.9%-11.2%) for artemether-lumefantrine ( $P < .05$  for all pairwise comparisons). When only recrudescence treatment failures were considered, the risks of failure were 14.1% (95% CI, 10.3%-19.2%), 4.6% (95% CI, 2.5%-8.3%), and 1.0% (95% CI, 0.3%-4.0%) for the same order of study drugs, respectively ( $P \leq .008$  for all pairwise comparisons, except amodiaquine plus artesunate vs artemether-lumefantrine,  $P = .05$ ). There were no deaths or cases of severe malaria. Significant reductions in anemia (9.3% [95% CI, 7.0%-12.0%] at enrollment vs 0.6% [95% CI, 0.1%-2.2%] during the last 2 months of follow-up;  $P < .001$ ) and asymptomatic parasitemia (18.6% [95% CI, 15.5%-22.1%] at enrollment vs 2.3% [95% CI, 1.5%-3.5%] during the last 2 months of follow-up;  $P < .001$ ) were observed according to routine testing.

**Conclusions** Artemether-lumefantrine was the most efficacious treatment for uncomplicated malaria in the study population. With all study regimens, the provision of prompt and reasonably effective facility-based treatment was associated with good outcomes in long-term health measures.

**Trial Registration** isrctn.org Identifier: ISRCTN37517549

JAMA. 2007;297:2210-2219

www.jama.com

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remaining 2 ACTs recommended by WHO, artemether-lumefantrine and amodiaquine + artesunate, have recently been adopted as first-line therapy by many countries in sub-Saharan Africa. However, because of limited supplies, difficulties in distribution, and economic constraints, the availability of ACTs remains limited and the majority of malaria episodes in Africa are not yet treated with these new regimens.

The WHO also recommends amodiaquine + sulfadoxine-pyrimethamine, a combination of 2 older drugs, as an alternative to ACT for the treatment of falciparum malaria, in which both drugs remain effective. Amodiaquine + sulfadoxine-pyrimethamine has shown surprisingly good efficacy in Ugandan studies despite known resistance to both components of the combination.<sup>7</sup> This regimen benefits from long half-lives of both drugs, providing improved protection against recurrent infection after therapy compared with amodiaquine + artesunate in Uganda.<sup>8</sup> However, the effectiveness of amodiaquine + sulfadoxine-pyrimethamine was poor in a recent study in Tanzania, and increasing resistance to each component drug may jeopardize this regimen.<sup>9</sup>

Antimalarial drug efficacy is most typically studied by following outcomes for a specified period, most commonly 28 days, after treatment for a single episode of malaria. However, recrudescence infections may occur more than a month after therapy, and different treatments may have varied effects on the likelihood of recrudescence and new infections after therapy. An improved means of comparing the overall efficacies of antimalarial regimens is the use of a longitudinal study format, whereby different therapies are compared throughout the course of multiple treatments for malaria.<sup>5</sup> Longitudinal studies also offer the advantage of measuring the incidence of malaria and evaluating long-term health outcomes.

To critically evaluate the 3 leading available combination regimens, we established a cohort of children repre-

sentative of an urban community in Kampala, Uganda, and compared the efficacy, safety, and tolerability against uncomplicated falciparum malaria of amodiaquine + sulfadoxine-pyrimethamine, amodiaquine + artesunate, and artemether-lumefantrine during an extended follow-up.

## METHODS

### Study Area and Recruitment of Cohort

The study was conducted between November 2004 and June 2006 in the Mulago III parish of Kampala, where malaria is mesoendemic, occurring throughout the year, with peaks during 2 rainy seasons. Before the onset of the study, a census project was carried out in the Mulago III parish from July to October 2004 to generate a sampling frame of households with appropriately aged children for recruitment and to gather basic demographic information about the target population.<sup>10</sup> All children from randomly selected households were screened for enrollment.

Enrollment occurred between November 2004 and April 2005. Study physicians recruited children if they fulfilled all the following eligibility criteria: aged 1 to 10 years, agreement to come to the study clinic for any febrile episode or illness, agreement to avoid medications administered outside the study, agreement to remain in Kampala during the study period, no known adverse reactions to the study medications, weight 10 kg or more, absence of severe malnutrition or known serious chronic disease, absence of life-threatening screening laboratory results, and willingness of parent or guardian to provide written informed consent. To allow evaluation of episodes of malaria diagnosed only during the period of observation and minimize the effect of antimalarial medications taken before enrollment in the study, children with symptomatic malaria on the day of screening were treated with quinine and enrolled only after documentation of a negative blood smear result 7 days after initiation of therapy.

The study received ethical approval from the Uganda National Council of Science and Technology, the Makerere University Research and Ethics Committee, and the University of California, San Francisco Committee on Human Research.

### Follow-up of Study Participants

Parents and guardians of study participants were asked to bring their children to the study clinic for all medical care. The study clinic was open daily from 8 AM to 5 PM, and after-hours care was available. Participants who presented to the study clinic with new medical problems underwent a standardized evaluation. Malaria was diagnosed if a child had complicated malaria (presence of severe malaria or danger signs)<sup>11</sup> or fever (documented tympanic temperature  $\geq 38.0^{\circ}\text{C}$  or history of fever in the previous 24 hours) and any parasitemia.

If no blood smear was performed after any 1-month interval, children underwent routine assessment and blood smear to ensure adherence with the study protocol and assess asymptomatic parasitemia. Routine complete blood cell count and alanine aminotransferase level tests were performed every 90 days. Medications with antimalarial activity were avoided for the treatment of nonmalaria illnesses when acceptable alternatives were available. Anthelmintics, iron sulfate, and vitamin A were routinely prescribed according to local Integrated Management of Childhood Illnesses guidelines of the Ugandan Ministry of Health.

### Treatment Allocation and Study Drug Administration

Study participants were randomly assigned to receive 1 of 3 oral antimalarial regimens at their first episode of uncomplicated malaria: amodiaquine plus sulfadoxine-pyrimethamine, amodiaquine plus artesunate, or artemether-lumefantrine. Participants received the same treatment regimen for all subsequent episodes of uncomplicated malaria diagnosed during the study period. The study medications were dosed

as follows: amodiaquine, 10 mg/kg on the first 2 days and then 5 mg/kg on the third day; sulfadoxine-pyrimethamine, sulfadoxine, 25 mg/kg, and pyrimethamine, 1.25 mg/kg as a single dose on the first day; artesunate, 4 mg/kg on all 3 days; and artemether-lumefantrine, 20 120-mg tablets given twice a day for 3 days according to weight: 5 through 14 kg, 1 tablet per dose; 15 through 24 kg, 2 tablets per dose; and 25 through 34 kg, 3 tablets per dose.

A randomization list was computer generated with variable blocks of 3, 6, and 9 by an off-site investigator. Sequentially numbered, sealed envelopes containing the treatment group assignments were prepared from the randomization list. The study nurse assigned treatment numbers sequentially and allocated treatment by opening the envelope corresponding to the treatment number. Study medications were administered by the nurses according to weight-based guidelines for administration of fractions of tablets modified from WHO recommendations. Participants in the amodiaquine + sulfadoxine-pyrimethamine and amodiaquine + artesunate groups also received placebo tablets, administered in the evening during 3 days, dosed similarly to weight-based guidelines for artemether-lumefantrine.

Administration of each first daily dose of medication was directly observed by the study nurses, and each second daily dose of medication or placebo was given to the participant's parent or guardian to administer at home in the evening. After treatment in the study clinic, patients were observed for 30 minutes and the dose was readministered if vomiting occurred. Patients who vomited persistently were referred for treatment with parenteral quinine. Participants with severe malaria or danger signs were also treated with quinine.

All study personnel involved in outcome assessment were blinded to treatment allocation. Study participants and their caregivers were not informed of their assigned treatment regimens; how-

ever, because the study drugs were not identical in appearance and taste, the study was considered single-blind.

### **Malaria Follow-up and Outcome Classification**

Participants diagnosed with malaria were asked to return on days 1, 2, 3, 7, 14, and 28 or any other day they felt ill. Follow-up evaluation consisted of a standardized medical history-taking and physical examination. Blood was obtained by finger prick for thick blood smears and storage on filter paper on all follow-up days, except day 1. A complete blood cell count and measurement of alanine aminotransferase were performed on the initial day of malaria diagnosis and on day 14.

Treatment outcomes were classified according to 2005 WHO guidelines as early treatment failure (complicated malaria or failure to adequately respond to therapy on days 0-3), late clinical failure (complicated malaria or fever and parasitemia on days 4-28, without previously meeting criteria for early treatment failure or late parasitological failure), late parasitological failure (asymptomatic parasitemia on days 7-28, without previously meeting criteria for early treatment or late clinical failure), and adequate clinical and parasitological response (absence of parasitemia on day 28, without previously meeting criteria for early treatment, late clinical, or late parasitological failure).<sup>12</sup> Patients with early treatment failure or late clinical failure within 14 days of initiation of therapy were treated with quinine, beginning a new 28-day follow-up schedule. Any case of symptomatic malaria diagnosed more than 14 days after a previous episode was considered a new event (for treatment purposes) and managed according to the protocol described above.

### **Adverse Event Monitoring**

Safety outcomes included risk of serious adverse events and risk of events of moderate or greater severity. Adverse event monitoring began with the first episode of uncomplicated ma-

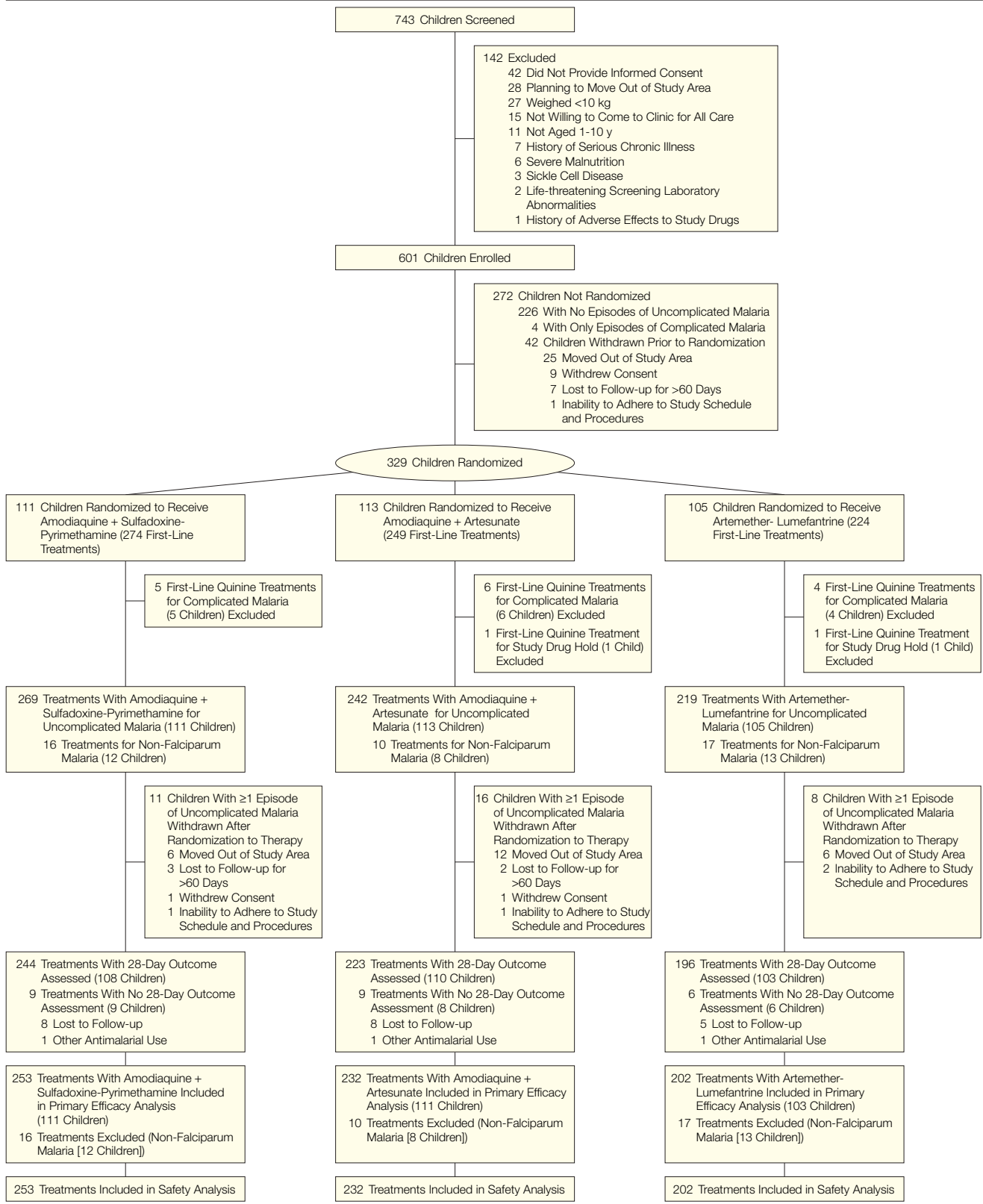
laria treated with study medications and continued for the remainder of the follow-up period, with assessment for new or worsening events at each visit. An adverse event was defined as any untoward medical occurrence, irrespective of its suspected relationship to the study medications, according to International Conference on Harmonisation guidelines. If a participant experienced a serious adverse event, he or she was not offered study treatment until the relationship of the event to the study medications could be established. If the event was deemed unrelated or only possibly related to the study medications, participation in the study was continued.

### **Laboratory Methods**

Thick and thin blood smears were stained with 2% Giemsa for 30 minutes. Parasite density was estimated by counting the number of asexual parasites per 200 white blood cells and calculating parasites per microliter, assuming a white blood cell count of 8000/ $\mu$ L.<sup>11</sup> A smear was judged to be negative if no parasites were seen after review of 100 high-powered fields. The diagnosis and management of malaria was based on initial readings of blood smears. Final microscopy results were based on a rigorous quality-control system that included rereading of all blood smears by a second microscopist and resolution of any discrepancies between the first and second readings by a third microscopist. No patients diagnosed with uncomplicated malaria based on the initial blood smear reading were considered not to have a positive blood smear result after the quality-control readings.

For episodes of recurrent parasitemia more than 3 days after the initiation of therapy, DNA was isolated from filter paper samples with chelex, and paired samples were genotyped in a stepwise fashion with *m*sp-2, *m*sp-1, and 4 microsatellites.<sup>13</sup> If, for any of the 6 loci, an allele was not shared between day 0 and the day of recurrence, the infection was classified as a new infection. If at least 1 allele was shared

**Figure 1. Trial Profile**



between day 0 and the day of recurrence at all 6 loci, the infection was classified as a recrudescence.

### Statistical Analysis

The sample size was calculated as follows. Assuming that 75% of participants would have at least 1 episode of malaria, the incidence of malaria treatments would be 2.75 per person-year in the reference group, and at a 10% attrition rate per year, it was estimated that 600 children would have to be followed up for 3 years to detect a 17% or greater difference in the incidence of malaria treatments (80% power, 2-sided  $\alpha$  level of .05).

An interim analysis was performed after approximately half the projected person-time was accrued (without adjustment). Stopping guidelines for the primary efficacy outcome in the study protocol recommended dropping the amodiaquine + artesunate or artemether-lumefantrine treatment arms only if they were found to be inferior to the amodiaquine + sulfadoxine-pyrimethamine treatment arm, given the lower cost of this regimen. The study protocol also proposed that the data and safety monitoring board not be strictly bound by prespecified criteria, because of the complexity of the tradeoffs between safety, efficacy, and costs and the possibility that new information would change considerations.

According to the results of the interim analysis, the data and safety monitoring board recommended that the amodiaquine + sulfadoxine-pyrimethamine treatment arm be dropped and the results of the study be presented early, given their potential public health implications and rapidly changing antimalarial therapy in Uganda and other African countries. However, the stopping of randomized trials early because of differences in treatment efficacy may lead to bias, resulting in overestimation of true differences between treatment arms.<sup>14</sup>

Data were double entered in Access (Microsoft Corporation, Redmond, Wash), and statistical analysis was per-

formed with Stata, version 8 (Stata-Corp, College Station, Tex). Efficacy and safety data were evaluated with an intention-to-treat analysis including all patients with falciparum malaria who were randomized to study drug therapy. Primary efficacy outcomes included 28-day risk for recurrent parasitemia (early treatment, late clinical, or late parasitological failure), both unadjusted and adjusted by genotyping to distinguish recrudescence and new infection. Risks of treatment failure were estimated with the Kaplan-Meier product limit formula.

Data were censored for patients who did not complete follow-up or were reinfected with non-falciparum species and for new infections caused by falciparum malaria according to outcomes adjusted by genotyping. Pairwise comparisons of treatment efficacy for individual episodes of malaria at 28 days of follow-up were made with a Cox proportional hazards model, with adjustment for repeated measures in the same patient. Pairwise comparisons of categorical variables were made with generalized estimating equations, with adjustment for repeated measures in the same patient by using exchangeable correlation and robust standard errors. Comparisons for the incidence of malaria treatments were made with a negative binomial regression model, with assigned treatment groups as covariates, exposure reflected by the time at risk after the first treatment with the study drug, and adjustment for correlations between children living in the same household.  $P < .05$  was considered statistically significant.

## RESULTS

### Trial Profile

A total of 743 children were screened for study participation, of whom 142 did not meet eligibility criteria (FIGURE 1). We enrolled 601 children from 323 households during a 6-month period and followed them up for 13 to 19 months.

At enrollment, sleeping under any bed net was reported for 258 (43%) children, and sleeping under an

insecticide-treated bed net was reported for 72 (12%). A total of 77 participants enrolled in the study were withdrawn before the end of the follow-up period for the following reasons: movement out of the study area ( $n=49$ ), inability to locate study participants for more than 60 consecutive days ( $n=12$ ), withdrawal of informed consent ( $n=11$ ), and inability to comply with the study schedule and procedures ( $n=5$ ). The total period of observation was 741 person-years, which covered 93% of the potential follow-up time. Of the 77 participants who were prematurely withdrawn from the study, 35 (45%) had developed at least 1 episode of uncomplicated malaria and were randomized to therapy before withdrawal.

Of the 601 children enrolled in the study, 329 children had at least 1 episode of uncomplicated malaria and were randomized to 1 of the 3 treatment arms (Figure 1). Among children randomized to study medication, 15 quinine treatments were given for complicated malaria, 2 quinine treatments were given for uncomplicated malaria when patients were on study treatment hold after potential drug-related adverse events, and 43 study medication treatments were given for uncomplicated non-falciparum malaria. In the comparative analysis of treatment efficacy, we included the remaining 687 study medication treatments given for uncomplicated falciparum malaria (Figure 1).

### Treatment Outcomes for Individual Episodes of Malaria

The baseline characteristics of episodes of uncomplicated falciparum malaria were similar with respect to patient age, temperature, parasite density, and hemoglobin among the 3 treatment arms (TABLE 1).

Treatment outcomes after 28 days of follow-up are presented in TABLE 2 and TABLE 3. Complete treatment outcomes were classified in more than 96% of cases, with outcomes not classified because of missed follow-up visits in 21 cases and antimalarial use outside the study protocol in 3 cases. Early treat-

ment failures within 3 days of diagnosis were uncommon (<1% of treatments) in the ACT treatment groups and occurred in 8 of 253 (3.2%) children treated with amodiaquine + sulfadoxine-pyrimethamine. Parasite clearance was clearly superior in the ACT treatment groups, with less than 3% of cases associated with a positive blood smear result on day 2 compared with 48% in the amodiaquine + sulfadoxine-pyrimethamine group ( $P < .001$ , TABLE 4). More rapid parasite clearance in the ACT treatment groups did not translate into a clear clinical benefit, however, because there was no consistent difference in the pattern of fever clearance between the 3 treatment groups (Table 4).

The most informative measures of treatment efficacy were estimates of treatment failure after 28 days of follow-up, both unadjusted and adjusted by genotyping (Table 3). There was a clear rank order of treatment efficacy among the 3 treatment groups. The risk of failure unadjusted by genotyping (including recrudescences and new infections) was 26.1% (95% confidence interval [CI], 21.1%-32.1%) with amodiaquine + sulfadoxine-pyrimethamine, 17.4% (95% CI, 13.1%-23.1%) with amodiaquine + artesunate, and 6.7% (95% CI, 3.9%-11.2%) with artemether-lumefantrine ( $P < .05$  for all

**Table 1.** Baseline Characteristics of All Uncomplicated Malaria Episodes Due to *Plasmodium falciparum* Treated With Study Drugs

| Characteristic  | Treatment Group                                   |                                    |                                   |
|---|---|------------------------------------|-----------------------------------|
|   | Amodiaquine + Sulfadoxine-Pyrimethamine (n = 253) | Amodiaquine + Artesunate (n = 232) | Artemether-Lumefantrine (n = 202) |
| Patient age, mean (SD), y                                     | 6.4 (2.5)   | 6.0 (2.5)                          | 6.2 (2.7)                         |
| Temperature, mean (SD), °C                                    | 37.9 (1.2)  | 37.9 (1.2)                         | 37.9 (1.2)                        |
| Parasite density, geometric mean, per $\mu$ L                 | 13 170  | 12 743                             | 10 939                            |
| Hemoglobin, mean (SD), g/dL                                   | 11.6 (1.3)  | 11.5 (1.4)                         | 11.5 (1.2)                        |
| Mixed infection with other <i>Plasmodium</i> species, No. (%) | 4 (1.6)   | 0                                  | 8 (4.0)                           |

**Table 2.** Treatment Outcomes After 28 Days for Episodes of Uncomplicated Falciparum Malaria

| Treatment Outcome                                  | Treatment Group, No. (%)                          |                                    |                                   |
|--|---|------------------------------------|-----------------------------------|
|  | Amodiaquine + Sulfadoxine-Pyrimethamine (n = 253) | Amodiaquine + Artesunate (n = 232) | Artemether-Lumefantrine (n = 202) |
| No treatment outcome                               | 9 (3.6)   | 9 (3.9)                            | 6 (3.0)                           |
| Lost to follow-up                                  | 8   | 8                                  | 5                                 |
| Other antimalarial use                             | 1   | 1                                  | 1                                 |
| Recurrent malaria caused by non-falciparum species | 0   | 0                                  | 3 (1.5)                           |
| Early treatment failure                            | 8 (3.2)   | 2 (0.9)                            | 1 (0.5)                           |
| Late clinical failure                              | 31 (12.3)   | 24 (10.3)                          | 5 (2.5)                           |
| Recrudescence                                      | 13  | 6                                  | 0                                 |
| New infection                                      | 18  | 17                                 | 5                                 |
| Genotyping unsuccessful                            | 0   | 1                                  | 0                                 |
| Late parasitological failure                       | 25 (9.9)  | 13 (5.6)                           | 7 (3.5)                           |
| Recrudescence                                      | 13  | 2                                  | 1                                 |
| New infection                                      | 12  | 11                                 | 6                                 |
| Adequate clinical and parasitological response     | 180 (71.1)  | 184 (79.3)                         | 180 (89.1)                        |

**Table 3.** Comparative Efficacies at 28 Days for Treatment of Uncomplicated Falciparum Malaria

|   | Risk of Treatment Failure, % (95% Confidence Interval) | Hazard Ratio (95% Confidence Interval)* | P Value* |
|---|--|---|----------|
| Unadjusted by genotyping†   |  |   |          |
| Amodiaquine + sulfadoxine-pyrimethamine vs amodiaquine + artesunate | 26.1 (21.1-32.1) vs 17.4 (13.1-23.1)                   | 1.58 (1.01-2.47)                        | .04      |
| Amodiaquine + sulfadoxine-pyrimethamine vs artemether-lumefantrine  | 26.1 (21.1-32.1) vs 6.7 (3.9-11.2)                     | 4.38 (1.99-9.63)                        | <.001    |
| Amodiaquine + artesunate vs artemether-lumefantrine                 | 17.4 (13.1-23.1) vs 6.7 (3.9-11.2)                     | 2.77 (1.22-6.30)                        | .02      |
| Adjusted by genotyping‡   |  |   |          |
| Amodiaquine + sulfadoxine-pyrimethamine vs amodiaquine + artesunate | 14.1 (10.3-19.2) vs 4.6 (2.5-8.3)                      | 3.21 (1.36-7.59)                        | .008     |
| Amodiaquine + sulfadoxine-pyrimethamine vs artemether-lumefantrine  | 14.1 (10.3-19.2) vs 1.0 (0.3-4.0)                      | 14.7 (3.59-59.9)                        | <.001    |
| Amodiaquine + artesunate vs artemether-lumefantrine                 | 4.6 (2.5-8.3) vs 1.0 (0.3-4.0)                         | 4.56 (0.99-21.0)                        | .05      |

\*Adjusted for repeated measures in the same patient.

†Treatment failure defined as any early treatment, late clinical, or late parasitological failure; episodes with no outcomes and recurrent malaria caused by non-falciparum species censored.

‡Treatment failure defined as any early treatment failure and only late clinical or late parasitological failure caused by recrudescence; episodes with no outcome, recurrent malaria caused by non-falciparum species, and new infections censored.

pairwise comparisons; Table 3). When only treatment failures caused by recrudescence parasites were considered, the risks of failure were 14.1% (95% CI, 10.3%-19.2%) with amodiaquine + sulfadoxine-pyrimethamine, 4.6% (95% CI, 2.5%-8.3%) with amodiaquine + artesunate, and 1.0% (95% CI, 0.3%-4.0%) with artemether-lumefantrine ( $P \leq .008$  for all pairwise comparisons, with the exception of amodiaquine + artesunate vs artemether-lumefantrine,  $P = .05$ ).

Given the longitudinal study design, we were able to assess the risk of recurrent symptomatic malaria after an extended period of follow-up. In the amodiaquine + sulfadoxine-pyrimethamine treatment group, 4 additional cases of recurrent malaria caused by recrudescence were identified after 28 days of follow-up. All late failures had asymp-

tomatic parasitemia on day 28, which progressed to symptomatic malaria between days 29 and 34. Recurrent malaria caused by recrudescence occurred after 28 days once in the amodiaquine + artesunate treatment group (on day 42) and twice in the artemether-lumefantrine treatment group (on days 37 and 42). The cumulative risk of recurrent malaria caused by recrudescence at 63 days of follow-up was 12.2%, 4.7%, and 1.8% in the amodiaquine + sulfadoxine-pyrimethamine, amodiaquine + artesunate, and artemether-lumefantrine treatment groups, respectively (FIGURE 2).

Different antimalarial therapies may vary in the risk of new infection after therapy because of different posttreatment prophylactic effects of the drugs. New infections were first identified 14, 21, and 23 days after initiation of therapy

in the amodiaquine + artesunate, amodiaquine + sulfadoxine-pyrimethamine, and artemether-lumefantrine treatment groups, respectively (Figure 2). The cumulative risk of new infection generally increased at a constant rate after therapy in the 3 treatment arms. There was no significant difference in the rate of new infection among the 3 treatment arms, although there was a trend toward a lower rate of new infection in the artemether-lumefantrine treatment group.

Secondary outcomes after treatment for individual episodes of malaria included gametocyte carriage and changes in hemoglobin levels. The prevalence of gametocytemia during the 14 days after initiation of therapy was lowest in the artemether-lumefantrine treatment group (Table 4). Changes in hemoglobin levels from day 0 to day 14 were similar among the 3 treatment arms (Table 4); only 12% of children were anemic (hemoglobin  $< 10$  g/dL) on the day malaria was diagnosed.

### Safety and Tolerability of Study Drugs

All the study regimens appeared to be safe and generally well tolerated. In the first 14 days after treatment with study medications, anorexia and weakness occurred more commonly in children treated with amodiaquine + sulfadoxine-pyrimethamine than those receiving amodiaquine + artesunate or artemether-lumefantrine (Table 4). A total of 45 serious adverse events were reported in 38 patients. Seizures were most commonly reported, with 18 episodes in 14 patients. The majority of seizures (78%) occurred in association with fever; 9 were classified as unrelated and 9 as possibly related to study medications. Elevation of liver enzyme levels occurred in 7 patients, all with causes other than study medications diagnosed (6 viral hepatitis and 1 *Salmonella* bacteremia). The other serious adverse events were attributable to illnesses other than malaria. No serious adverse event was considered to be probably or definitely related to the study medications.

**Table 4.** Secondary Outcomes for Episodes of Uncomplicated *Falciparum* Malaria

| Outcome  | Treatment Group                                   |                                    |                                   |
|--|---|------------------------------------|-----------------------------------|
|  | Amodiaquine + Sulfadoxine-Pyrimethamine (n = 253) | Amodiaquine + Artesunate (n = 232) | Artemether-Lumefantrine (n = 202) |
| Fever clearance, No. (%) <sup>*</sup>              |   |                                    |                                   |
| Fever on day 1†‡                                   | 101 (40)  | 135 (59)                           | 128 (63)                          |
| Fever on day 2†                                    | 36 (22)   | 29 (13)                            | 32 (16)                           |
| Fever on day 3                                     | 9 (3.7)   | 11 (4.8)                           | 6 (3.0)                           |
| Parasite clearance, No. (%)                        |   |                                    |                                   |
| Parasitemia on day 2†‡                             | 120 (48)  | 5 (2.2)                            | 6 (3.0)                           |
| Parasitemia on day 3†‡                             | 15 (6.2)  | 0                                  | 0                                 |
| Gametocytes by day, No. (%)                        |   |                                    |                                   |
| 0  | 19 (7.5)  | 21 (9.1)                           | 13 (6.4)                          |
| 2  | 17 (6.8)  | 19 (8.3)                           | 5 (2.5)                           |
| 3‡   | 25 (10)   | 15 (6.6)                           | 4 (2.0)                           |
| 4-14†‡   | 33 (14)   | 14 (6.1)                           | 7 (3.5)                           |
| Change in hemoglobin, mean (SD), g/dL§             | 0.16 (1.03)                                       | -0.03 (1.10)                       | 0.09 (1.01)                       |
| Adverse events of any severity, days 1-14, No. (%) |   |                                    |                                   |
| Anorexia†‡   | 82 (32)   | 56 (24)                            | 39 (19)                           |
| Cough  | 78 (31)   | 66 (29)                            | 59 (29)                           |
| Weakness†‡   | 64 (25)   | 37 (16)                            | 29 (14)                           |
| Abdominal pain                                     | 41 (17)   | 40 (19)                            | 37 (20)                           |
| Vomiting   | 40 (16)   | 32 (13)                            | 26 (13)                           |
| Diarrhea   | 23 (9)  | 21 (9)                             | 19 (9)                            |
| Pruritus   | 24 (9)  | 25 (11)                            | 23 (11)                           |
| Any severe adverse event                           | 16 (6)  | 15 (6)                             | 14 (7)                            |

<sup>\*</sup>Subjective fever during previous 24 hours or temperature  $\geq 38.0^\circ\text{C}$ .

†Amodiaquine + sulfadoxine-pyrimethamine vs amodiaquine + artesunate,  $P < .05$  (adjusted for repeated measures in the same patient).

‡Amodiaquine + sulfadoxine-pyrimethamine vs artemether-lumefantrine,  $P < .05$  (adjusted for repeated measures in the same patient).

§Change from day 0 to day 14 or day of clinical failure.

||Amodiaquine + artesunate vs artemether-lumefantrine,  $P < .05$  (adjusted for repeated measures in the same patient).

### Longitudinal Outcomes

Given the longitudinal study design, we measured the incidence of malaria and examined long-term malaria-related outcomes. Considering all treatments for malaria after the first treatment with study drugs, children randomized to the artemether-lumefantrine group had a 32% reduction (95% CI, 5%-51%) in the incidence of malaria treatments compared with children randomized to amodiaquine + sulfadoxine-pyrimethamine (1.24 vs 1.74;  $P=.02$ ). Children randomized to the amodiaquine + artesunate treatment group had a non-significant 24% reduction (95% CI, -5% to 45%) in the incidence of malaria treatments compared with children randomized to amodiaquine + sulfadoxine-pyrimethamine (1.34 vs 1.74;  $P=.10$ ). There was no significant difference in the incidence of malaria treatment for children randomized to the 2 ACT regimens (1.24 vs 1.34;  $P=.53$ ). The overall health of the children in this cohort was excellent. There were no deaths and no episodes of malaria that met WHO criteria for severe malaria. There were 19 episodes of malaria that were considered complicated and

treated with quinine for the following reasons: single seizures ( $n=12$ ), hyperparasitemia defined as parasite density greater than 500 000/ $\mu\text{L}$  ( $n=3$ ), inability to sit up or stand ( $n=2$ ), persistent vomiting ( $n=1$ ), and lethargy ( $n=1$ ). There was a marked reduction in the prevalence of asymptomatic parasitemia and anemia (hemoglobin  $<10$  g/dL) in the cohort throughout the study (FIGURE 3). At enrollment, 104 of 559 (18.6%) asymptomatic children had a positive blood smear result compared with 22 of 962 (2.3%) asymptomatic children assessed during the last 2 months of follow-up ( $P<.001$ ). At enrollment, 52 of 559 (9.3%) asymptomatic children were anemic compared with 2 of 331 (0.6%) asymptomatic children tested during the last 2 months of follow-up ( $P<.001$ ).

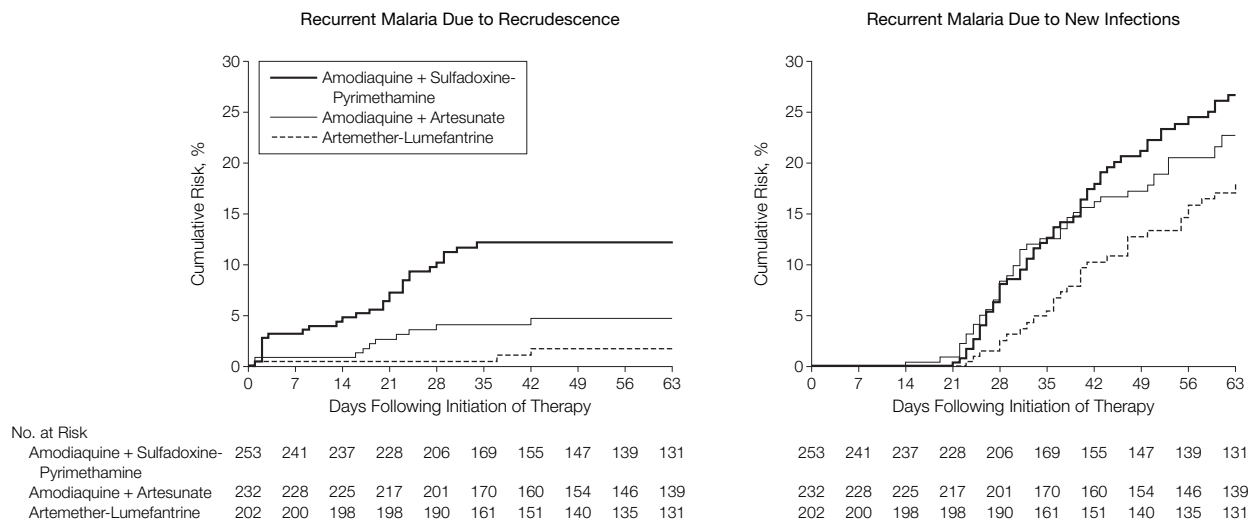
### COMMENT

In this randomized clinical trial, there was a rank order in efficacy between the 3 treatment groups, with artemether-lumefantrine providing the best treatment outcomes, followed by amodiaquine + artesunate then amodiaquine

+ sulfadoxine-pyrimethamine. Only 1 early treatment failure and 1 recrudescence occurred among 202 patients treated with artemether-lumefantrine after 28 days of follow-up. Thus, of combination regimens available to treat uncomplicated malaria in Kampala, artemether-lumefantrine is the superior regimen. The overall health of the children in the cohort was excellent, regardless of the assigned treatment group. No episodes of malaria were classified as severe, and no deaths occurred. In addition, the prevalence of asymptomatic parasitemia and anemia decreased significantly during the study. The long-term health benefits observed in our cohort suggest that provision of health facility-based care and treatment of microscopically confirmed malaria with effective combination regimens may offer important advantages over the widespread strategy of community-based management of febrile illnesses in Africa.

The combination of amodiaquine + sulfadoxine-pyrimethamine has been well studied in Uganda.<sup>5,7,8</sup> Despite widespread resistance to the individual agents, this inexpensive combi-

**Figure 2.** Cumulative Risk of Recurrent Malaria Due to Recrudescence and New Infections Calculated With the Kaplan-Meier Product Limit Formula



Pairwise comparisons of survival curves using Cox proportional hazards model with adjustment for repeated measures in the same patient. Recrudescence: amodiaquine + sulfadoxine-pyrimethamine vs amodiaquine + artesunate ( $P=.02$ ), amodiaquine + sulfadoxine-pyrimethamine vs artemether-lumefantrine ( $P<.001$ ), amodiaquine + artesunate vs artemether-lumefantrine ( $P=.09$ ). New infections: amodiaquine + sulfadoxine-pyrimethamine vs amodiaquine + artesunate ( $P=.60$ ), amodiaquine + sulfadoxine-pyrimethamine vs artemether-lumefantrine ( $P=.19$ ), amodiaquine + artesunate vs artemether-lumefantrine ( $P=.40$ ).

nation has proven to be surprisingly efficacious. Indeed, the efficacy of amodiaquine + sulfadoxine-pyrimethamine was comparable, or even superior, to ACT regimens in several studies, in general because amodiaquine + sulfadoxine-pyrimethamine offered better protection than other regimens against new infections developing after therapy.<sup>5,7,8</sup> However, concerns have been raised about the safety, tolerability, and continued efficacy of amodiaquine + sulfadoxine-pyrimethamine. In Rwanda, where amodiaquine + sulfadoxine-pyrimethamine was adopted as first-line therapy for malaria in 2001, subsequent studies suggested that this regimen is failing and that amodiaquine-containing regimens are less well tolerated than alternative treatments.<sup>15,16</sup> In Muheza, Tanzania, where resistance to amodiaquine and sulfadoxine-pyrimethamine is common, the effectiveness of amodiaquine + sulfadoxine-pyrimethamine was poor.<sup>9</sup>

Our study suggests that the efficacy of amodiaquine + sulfadoxine-pyrimethamine is decreasing in Kampala; only 74% of children treated with amodiaquine + sulfadoxine-pyrimethamine had clinically successful outcomes at day 28 compared with 84% at the same site in 2002-2003.<sup>7</sup> Amodiaquine + sulfadoxine-pyrimethamine was inferior to the 2 ACT regimens

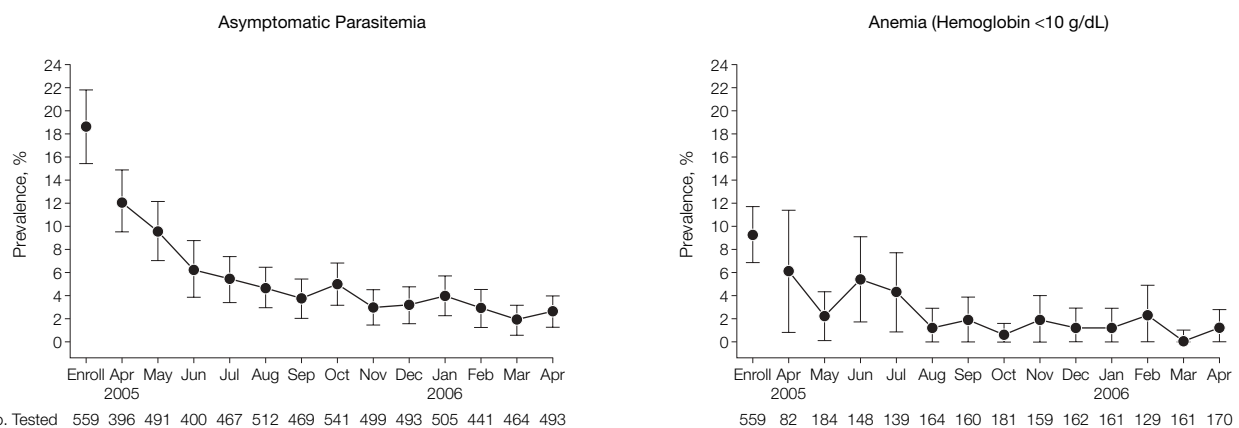
evaluated in this study. However, the efficacy of amodiaquine + sulfadoxine-pyrimethamine likely remains adequate in other parts of Africa. In Burkina Faso, where the efficacies of both amodiaquine and sulfadoxine-pyrimethamine are superior to those in East Africa, amodiaquine + sulfadoxine-pyrimethamine was superior to artemether-lumefantrine because of a decreased risk of new infections after therapy.<sup>17</sup> These data suggest that antimalarial regimens must be tailored according to local drug sensitivity patterns, as well as other factors such as safety, tolerability, cost, and availability.

All ACTs should not be considered equivalent regimens. The use of artemisinin monotherapy has been associated with high rates of late recrudescence,<sup>18</sup> so the success of an ACT regimen depends on the ability of the longer-acting partner drug to clear residual parasites. As resistance to a partner drug increases, an ACT regimen will increasingly fail, as evident from studies of chloroquine + artesunate in West Africa<sup>19</sup> and sulfadoxine-pyrimethamine + artesunate in East Africa.<sup>5,6</sup> A similar phenomenon may be occurring with amodiaquine + artesunate in East Africa, where increasing levels of resistance to amodiaquine were associated with worrisome failure rates with

amodiaquine + artesunate in this study and others.<sup>9</sup> In contrast, artemether-lumefantrine has the distinct advantage among available ACT regimens of containing a partner drug that has never been used as monotherapy. Therefore, unlike the situation with amodiaquine, parasite resistance to lumefantrine is unlikely. Results of this study and others confirm that artemether-lumefantrine offers the most effective available antimalarial regimen in East Africa, where drug resistance is the highest on the continent.<sup>9</sup>

Studies of antimalarial drug efficacy have typically focused on individual episodes of disease, with follow-up limited to 2 to 4 weeks. These short-term evaluations may not be adequate to estimate the true effect of drug resistance, particularly for longer-acting agents and more effective combination therapies.<sup>20,21</sup> Rather, important differences may be apparent only after extended evaluation. Recently, WHO has modified the suggested protocol for in vivo studies of antimalarial drug efficacy, recommending that patients be followed up for at least 28 days and that reappearance of parasites lead to a classification of failure regardless of whether clinical signs or symptoms recur.<sup>12</sup> Results from this study support the new WHO recommendations. All episodes of recurrent malaria caused by

**Figure 3.** Prevalence of Asymptomatic Parasitemia and Anemia at Enrollment (November 2004–April 2005) and at Monthly Intervals During Follow-up



Error bars indicate 95% confidence intervals.

recrudescence after amodiaquine + sulfadoxine-pyrimethamine therapy were identified as late clinical or parasitological failures by day 28. Extending follow-up to 42 days identified 1 additional episode of recurrent malaria caused by recrudescence in the amodiaquine + artesunate group and 2 additional episodes in the artemether-lumefantrine group. No episodes of malaria caused by recrudescence were identified beyond 42 days of follow-up. In addition, 39 of 45 (87%) children with late parasitological failures identified by day 28 went on to develop symptomatic malaria.

The excellent outcomes for all study participants suggest that even in the setting of suboptimal therapy, major advances in child health can be achieved by providing reasonably effective health facility-based treatment, ensuring good adherence, and targeting antimalarial therapy to microscopically confirmed cases of malaria. However, the results of this study should not be generalized to other epidemiologic settings. In rural areas, the incidence of malaria may be significantly higher than in the urban environment, and access to prompt high-quality care is often lacking.

In Uganda, many antimalarial treatments are administered presumptively, according only to the report of fever, often through the private sector. In addition, a program of home-based presumptive treatment of febrile children with antimalarial medications has been implemented, with plans to replace the older national regimen of chloroquine + sulfadoxine-pyrimethamine with ACTs. However, presumptive treatment of all fevers with ACTs will be expensive, limit the availability of drugs for treatment of confirmed malaria cases, lead to delays in treatment for nonmalaria illnesses, unnecessarily expose patients to potential adverse effects, and likely contribute to the development of drug resistance.

Considering the availability of resources such as light microscopy and

rapid diagnostic tests, as well as increasing funding through such programs as the Global Fund and President's Malaria Initiative, it seems that improved malaria management, with evaluation and diagnosis-based treatment for all febrile children, is a reasonable goal for Africa. Continued research into malaria diagnostics, optimal antimalarial regimens, sustainable methods of drug delivery, and integration of treatment with prevention strategies will be necessary to establish effective and sustainable malaria control policies.

**Author Contributions:** Dr Dorsey had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

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**Critical revision of the manuscript for important intellectual content:** Dorsey, Staedke, Clark, Njama-Meya, Dokomajilar, Kanya, Rosenthal.

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**Study supervision:** Dorsey, Staedke, Clark, Kanya, Rosenthal.

**Financial Disclosures:** None reported.

**Funding/Support:** The study received financial support from the National Institutes of Health (U01 AI052142) and the Doris Duke Charitable Foundation (Dr Rosenthal is a Doris Duke Charitable Foundation Distinguished Clinical Scientist).

**Role of the Sponsor:** The funding agencies had no role in the design and conduct of the study; collection, management, analysis, and interpretation of the data; and preparation, review, or approval of the manuscript.

**Acknowledgment:** We are grateful to all the parents and guardians for kindly giving their consent and the study participants for their cooperation. We thank all the members of the study team in Uganda.

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