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RESEARCH

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Superiority trial of intermittent treatment with dihydroartemisinin–piperaquine versus sulfadoxine–pyrimethamine for the prevention of malaria during pregnancy

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Abstract

Background Malaria in pregnancy is responsible for various adverse maternal and birth outcomes. The emerging resistance to sulfadoxine–pyrimethamine (SP) raises important concerns about its use for intermittent preventive treatment in pregnancy (IPTp) in Africa. This trial aimed to assess the efficacy and safety of IPTp with dihydroartemisinin–piperaquine (DP) as an alternative to IPTp with SP.

Results The double-blind, randomized, and controlled superiority trial was conducted between July 2020 and June 2021. A total of 250 women were enrolled and randomly assigned to receive SP ($n=125$) or DP ($n=125$). Two hundred and six (82.4%) participants that contributed to the outcomes were included in the modified intention-to-treat (ITT) analysis, while 84 participants that completed the three courses of the study drugs were included in the per protocol (PP) analysis. The ITT analysis results showed that the incidence of histopathologically confirmed placental malaria was nonsignificantly higher in the DP group compared with the SP group (62.5% vs. 51.1%, $P=0.098$). After adjusting for confounders, the risk of histopathologically confirmed placental malaria was also nonsignificantly higher in the DP group (Adjusted Relative Risk [RR] = 1.27, 95% CI 0.94–1.71) compared with the SP group. In contrast, the risk of a low APGAR score was significantly lower in the DP group (RR = 0.45, 95% CI 0.38–0.52) compared with the SP group. Also, the risk of a composite adverse birth outcome (low birth weight or preterm delivery or neonates small for the gestational age) was nonsignificantly lower in the DP group (Adjusted RR = 0.82, 95% CI 0.55–1.21) compared with the SP group. Both drugs were well tolerated, although nausea and vomiting occurred in a significant number of participants in the SP group.

Conclusions A three-course IPTp with DP was safe and was not found to be superior to IPTp with SP in the prevention of placental malaria. Although IPTp with DP was associated with a significant lower risk of low APGAR score and nonsignificant lower risks of other adverse birth outcomes compared with IPTp with SP.

Trial registration PACTR, PACTR202002644579177. Registered 20 February 2020, https://pactr.samrc.ac.za/TrialDisplay.aspx?TrialID=9753.

Keywords Dihydroartemisinin–piperaquine, Intermittent preventive treatment, Malaria, Pregnancy, Sulfadoxine–pyrimethamine

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Background

Malaria in pregnancy is responsible for various adverse outcomes such as maternal anemia, intrauterine growth restriction, stillbirth, premature delivery, low birth weight, neonatal morbidity, and mortality [1, 2]. Thus, the use of insecticide-treated bed nets coupled with intermittent preventive treatment in pregnancy (IPTp) with sulfadoxine–pyrimethamine (SP) in the second and third trimesters was recommended by WHO in malaria-endemic regions of sub-Saharan Africa [3]. However, the spread of resistance to SP in Africa is attributed to triple mutations at codons 51 (N51I), 59 (C59R) and 108 (S108N) of the *dihydrofolate reductase (dhfr)* gene and to quintuple mutations mediated by the triple *dhfr* mutant + double *dihydropteroate synthase (dhps)* mutant gene at codons 437 (A437G) and 540 (K540E) of the parasite [4, 5]. This raises important concerns about the use of SP for IPTp in Africa. A recent study reported the emergence of the dhps-431 V halotype all over Nigeria [6]. There is a knowledge gap on how long IPTp with SP will remain effective vis-a-vis this increasing level of resistance of *Plasmodium falciparum* to SP in Nigeria. Therefore, it is crucial to evaluate alternative antimalarial for IPTp in case of complete failure of SP.

Dihydroartemisinin–piperaquine (DP) is an artemisinin-based combination therapy (ACT) that could be a possible replacement for SP due to its many desirable characteristics. Dihydroartemisinin–piperaquine is tolerated well, has a high malaria parasite clearance capacity and the partner drug (piperaquine) provides at least 4 weeks of post-treatment prophylaxis [7, 8]. Intermittent preventive treatment in pregnancy trials comparing DP and SP that have been published to date have been conducted in East Africa where the prevalence and type of SP resistance markers differ from West Africa. In Tanzania, a trial that evaluated the efficacy of monthly IPTp with DP and SP found a significantly lower prevalence of placental malaria in IPTp with DP than IPTp with SP. In this study, the prevalence of composite adverse birth outcomes was not significant between both groups, although the prevalence of low birth weight was significantly lower in the DP group compared with the SP group [9]. In the Ugandan trials, the prevalence of histopathologically confirmed placental malaria was significantly lower in the DP group than in the SP group [10, 11]. While there were no significant differences between the groups in the risk of adverse drug events, the prevalence of adverse birth outcomes were nonsignificantly higher in the DP group than in the SP group [10, 11]. The Kenyan trial showed no significant difference between both groups in the prevalence of polymerase chain reaction and histopathologically confirmed placental malaria and adverse birth outcomes [12]. To our knowledge, this is the first study evaluating IPTp with DP as an alternative to IPTp with SP in West Africa. Therefore, this trial aimed to assess the efficacy and safety of IPTp with DP as an alternative to IPTp with SP.

Methods

Trial design and setting

A double-blind, randomized, and controlled superiority clinical trial comparing a three-course IPTp with SP, and a three-course IPTp with DP was conducted at a tertiary hospital in Maiduguri, Nigeria. The trial setting is a high transmission and mesoendemic area for malaria infections with emerging SP resistance [13, 14].

Sample size determination

A prevalence of acute placental malaria of 33.9% in the SP group on the basis of previous data was assumed to test the hypothesis that the use of IPTp with DP would be associated with a lower prevalence of histopathologically confirmed active and/or past placental malaria than that associated with SP. A calculated sample size of 100 would be required per group or a total of 200 for the study to have 80% power to show a 50% lower prevalence with DP, at a significance level of 0.05. Twenty percent was added to account for attrition and a total of 250 women were recruited for the study.

Inclusion criteria

Eligible participants were falciparum malaria rapid diagnostic test (RDT) and HIV-negative pregnant adolescents or women of at least 15 years of age (primigravid or multigravid) between 16 and 20 weeks of gestation, as confirmed by last menstrual period or ultrasound scan. Those with no current history of receiving any IPTp, and willing to provide informed written consent were also eligible. A complete list of the entry criteria is provided in the trial protocol (The Pan African Clinical Trials Registry Identifier: PACTR202002644579177), available at https://pactr.samrc.ac.za/TrialDisplay.aspx?TrialID=9753.

Exclusion criteria

Exclusion criteria were having a history of allergic reaction to sulfa drugs (SP or cotrimoxazole) or DP, having hemoglobin level < 7 g/dL, RDT-positive test for falciparum malaria, and history of receipt of antimalarials or antibiotics with antimalarial activity (cotrimoxazole, rifampin, doxycycline, clindamycin, tetracycline, erythromycin, azithromycin, chloramphenicol) in the past month. Others were women with sickle cell disease, women with high-risk pregnancies (presence of high blood pressure and diabetes), and HIV-positive women.

Randomization and masking

A pharmacist who was not part of the drug administration generated 25 blocks of equal size of 10 random numbers using a 1:1 allocation ratio to ensure that equal numbers of the participants were assigned to SP, serving as the control group or DP group, serving as the experimental group. He used the randomization sequence to package and label the trial agents. The study case report form marked with the randomization code was folded inside an opaque small envelope containing the appropriate study agents with the same code, which was finally placed in a big envelope representing each block. Another pharmacist was responsible for treatment assignments. The investigators and outcome assessors were also blinded for the group allocation. Each dose of SP (tablets of 500 mg of sulfadoxine and 25 mg of pyrimethamine [Amalar®, Elbe Pharma]) consisted of three tablets taken together on the first day and 3 tablets of placebo with the same appearance also taken together for additional 2 days; doses were administered at three times during the pregnancy. It was not possible to administer the three-dose regimen of each trial drug through directly observed therapy instead participants were instructed to take them at home once daily for 3 days. Each dose of DP (tablets of 40 mg of dihydroartemisinin and 320 mg of piperaquine [Ibasunate®, Elbe Pharma]) consisted of three tablets given once a day for three consecutive days; doses were also administered three times during the pregnancy. Participants who were assigned to the SP group or DP group received active trial agents at 16–20, 28, and 36 weeks. A case report form marked with the unique allocation code and with no information on the IPTp regimen participants received was used to collect the trial data.

Procedures

At enrollment, participants received a net treated with long-lasting insecticide with instructions to sleep under it regularly. Also, they picked up the coded small envelopes containing the trial agent from the big envelope and had blood samples collected for hemoglobin test. Routine visits were scheduled every four weeks, and blood samples were collected for routine laboratory testing when necessary. Measurements of alanine aminotransferase (ALT) levels were performed at enrollment and eight weeks afterward. Participants were encouraged to give birth at the trial hospital. At delivery, a standardized assessment was completed, including evaluation of the neonate for congenital anomalies, measurement of birth weight, determination of Appearance, Pulse, Grimace, Activity, and Respiration (APGAR) score, and collection of placental tissue for analyses.

Placental tissues were fixed in 10% buffer formalin and then processed and stained with modified Giemsa [15]. Placental malaria was diagnosed using the Rogerson criteria [16]. Histopathological slides were read in duplicate by two independent histopathologists, and the results were recorded on a standardized case-record form; any discrepant results were resolved by a third histopathologist. The histopathologists were unaware of both the assigned treatments and the previous malaria parasite test results.

Outcomes

The primary outcome was the incidence and risk of placental malaria defined as the histopathologically confirmed active and/or past placental malaria infection. A participant was determined to have active or chronic or past placental malaria infection if she had parasites or pigment in her placenta determined by histopathology of the placental tissue [16]. Secondary outcomes were the risks of adverse birth outcomes (spontaneous abortion [expulsion of a fetus before 28 weeks of gestation], stillbirth [delivery of a dead child after 28 weeks of gestation], low birth weight [< 2500 g], preterm delivery [birth at < 37 weeks of gestation], small for gestational age [birth weight below the 10th percentile], congenital anomaly (birth defects visible at birth or diagnosed by screening tests such as routine ultrasound examinations), low APGAR score at 5th minutes (< 7), and a composite of any of these birth outcomes), and the occurrences of adverse drug effects. For participants who gave birth to twins, the delivery outcomes were based on whether the outcome was present in either child or in the placenta. Measures of safety and side-effect profiles included the prevalence of elevated ALT levels (< 36 IU/L) eight weeks after the administration of trial agents and the occurrence of adverse effects after the initiation of trial agents through delivery.

Statistical analysis

Statistical analyses were performed with IBM-SPSS software, version 25. Analyses were performed using the modified intention-to-treat (ITT) for all participants that had at least one outcome and per protocol (PP) approach for all participants that completed the three courses IPTp with at least an outcome of the trial. Comparisons of proportions were performed using the chi-square test or Fisher's exact test (where appropriate). Continuous data were compared between the two treatment groups using the independent samples t test. Log binomial regression analyses were performed to obtain crude and adjusted relative risk (RR) values and corresponding 95% confidence intervals for binary outcomes. A p value of less than 0.05 was considered statistically significant.

Results

The trial was conducted from July 1, 2020 to June 30, 2021. A total of 323 women were screened and 250 were enrolled and underwent randomization with 125 participants each assigned to the SP group and DP group, respectively. A total of 206 participants (94 in the SP group vs. 112 in the DP group) that were followed through delivery were included in the ITT analysis, while 84 participants that completed the three courses of the trial agents (36 in the SP group vs. 48 in the DP group) were included in PP analysis as shown in Fig. 1.

In Table 1, the baseline characteristics were similar among the two treatment groups. The mean age at enrollment was 27.7 ± 6.1 years in the SP group and 27.3 ± 5.7 years in the DP group, and 57.4% and 64.3% of the participants were enrolled in the SP and DP groups, respectively, between 16 and 17 weeks of gestation. The common parity group was multigravidae (64.9% in the SP vs. 66.1% in the DP group), while 43.6% of the SP group and 38.4% of the DP group were anemic at enrollment. The mean ALT levels were 11.5 ± 6.9 IU/L and 12.0 ± 7.9 IU/L in the SP and DP groups, respectively.

The modified ITT analysis indicated a nonsignificant higher incidence of placental malaria in the DP group compared with the SP group (63.1% vs. 51.1%). Also, there was a nonsignificant higher risk of placental malaria in the DP group (RR = 1.29, 95% CI 0.96–1.73) compared with the SP group. This risk was slightly reduced after adjusting for confounders, although nonsignificant result was still maintained (Adjusted RR= 1.27, 95% CI 0.94-1.71). Subgroup analyses by gravidity

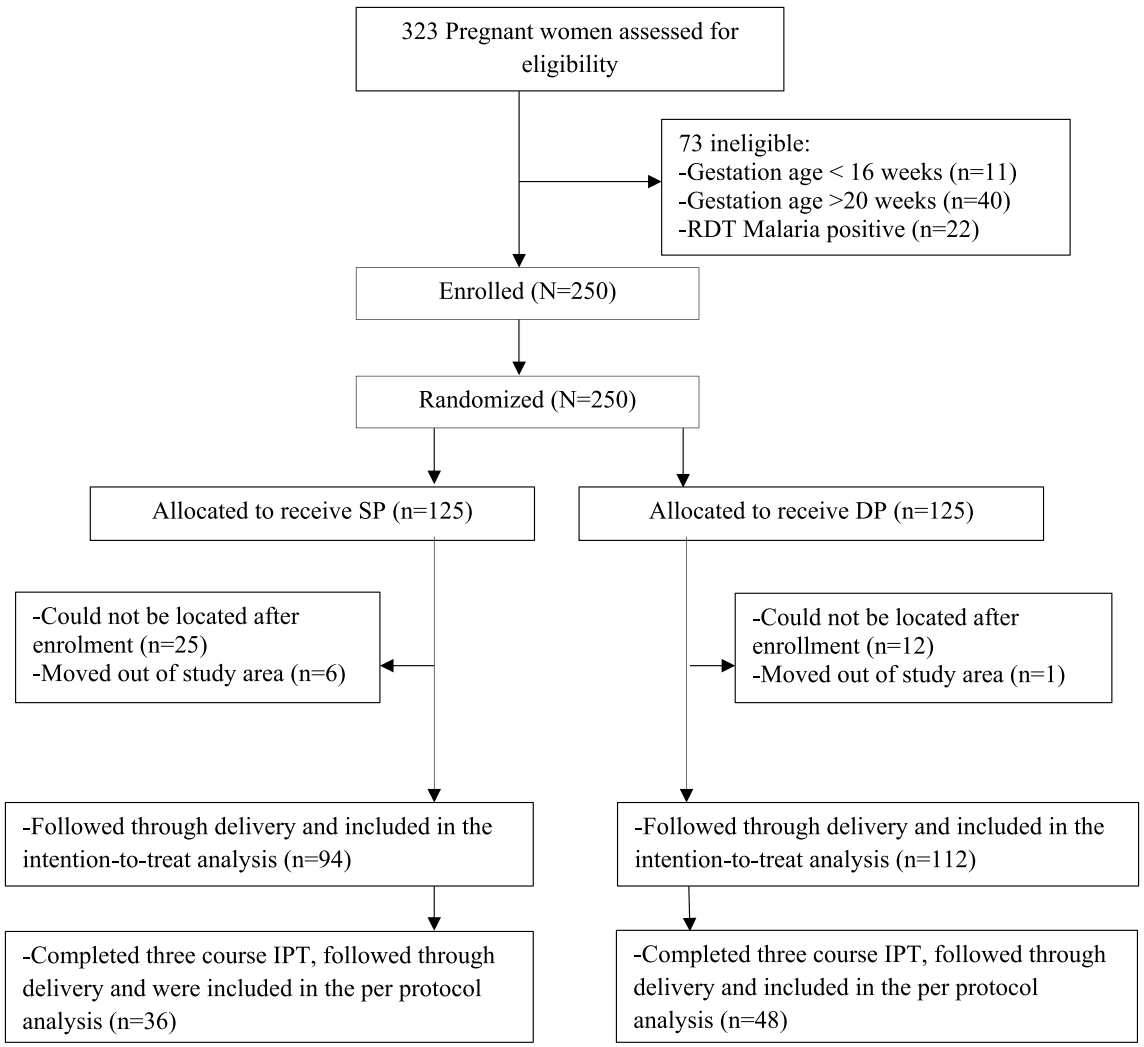

Fig. 1 CONSORT diagram of participant flow

Table 1 Baseline characteristics of the participants that were followed up through delivery ($N=206$)

Variables	Trial groups	
	SP $n=94$	DP $n=112$
Mean age (years)[a]	27.7 ± 6.1	27.3 ± 5.7
Age-group (years), n (%)[b]		
17–25	35 (37.2)	50 (44.6)
26–33	43 (46.7)	43 (38.4)
> 33	16 (17.1)	19 (17.0)
Mean gestational age in weeks[a]	17.7 ± 1.7	17.4 ± 1.6
Gestation age in weeks, n (%)[b]		
16–17	54 (57.4)	72 (64.3)
18–20	40 (42.6)	40 (35.7)
Gravidity, n (%)[b]		
Primigravidae	33 (35.1)	38 (33.9)
Multigravidae	61 (64.9)	74 (66.1)
Mean weight (kg)[a]	65.8 ± 14.8	63.6 ± 13.8
Mean height (m)[a]	1.59 ± 0.07	1.59 ± 0.08
BMI (kg/m^2), n (%)[b]		
< 18.5	6 (6.4)	10 (9.0)
18.5–24.9	40 (42.6)	52 (46.4)
> 24.9	48 (51.0)	50 (44.6)
Mean hemoglobin (g/dL)[a]	10.3 ± 1.4	10.4 ± 1.4
Anemia, n (%)[b]	41 (43.6)	43 (38.4)
Mean ALT level (IU/L)[a]	11.5 ± 6.9	12.0 ± 7.9
Elevated ALT level (> 36 IU/L), n (%)c[c]	1 (1.1)	3 (2.7)

ALT Alanine aminotransferase

[a] Independent samples *t* test

[b] Chi-square test

[c] Fisher's exact test

revealed a significant higher risk of placental malaria among the primigravidae in the DP group compared to primigravidae in the SP group. This significant risk was maintained even after adjusting for possible confounding variables (RR = 1.89, 95% CI 1.13–3.16). There was a nonsignificant reduced risk of spontaneous abortion in the DP group compared with the SP group (RR = 0.68, 95% CI 0.30–1.54). This reduced risk was significant in the primigravidae (RR = 0.45, 95% CI 0.35–0.58). Also, the risks of low birth weight (RR = 0.76, 95% CI 0.53–1.10), and composite adverse birth outcomes (RR = 0.83, 95% CI 0.57–1.21) were nonsignificantly lower in the DP group compared with the SP group. These reduced risks remained nonsignificant even after adjustment for possible confounders. Also, the risk of a low APGAR score was significantly lower in the DP group compared with the SP group (RR = 0.45, 95% CI 0.38–0.52). Both the primigravidae (RR = 0.45, 95% CI 0.35–0.58), and multigravidae (RR = 0.44, 95% CI 0.37–0.54) in the DP group maintained this significant lower risk. Conversely, the risks of preterm delivery (RR = 1.01, 95% CI 0.52–1.95), and small for gestational age (RR = 1.07, 95% CI 0.45–2.54) were, respectively, nonsignificantly slightly higher in the DP group compared to the SP group. However, the subgroup analyses by gravidity for the risks of preterm delivery and small for gestational age, respectively, resulted still in nonsignificant changes in the risk for either subgroup (Table 2).

In Table 3, the PP analysis results also showed no significantly higher incidence of placental malaria in the DP group compared with the SP group (56.2% vs. 52.8%, $P=0.752$). There was also no significantly slightly higher risk of placental malaria in the DP group (RR = 1.08, 95% CI 0.66–1.77) compared with the SP group. The subgroup analysis by gravidity revealed that in the DP group, both primigravidae (RR = 1.29, 95% CI 0.50–3.28), and multigravidae (RR = 1.02, 95% CI 0.57–1.82) also had nonsignificant higher risks of placental malaria compared to their counterparts in the SP group. The risk of low APGAR score was significantly lower in the DP group compared with the SP group (RR = 0.42, 95% CI 0.32–0.52). The multigravidae in the DP group showed a significant lower risk of low APGAR score compared to their counterpart in the SP group (RR = 0.43, 95% CI 0.32–0.59). Also, the risks of low birth weight (RR = 0.65, 95% CI 0.36–1.19), pre-term delivery (RR = 0.63, 95% CI 0.27–1.46), small for gestational age (RR = 0.63, 95% CI 0.27–1.46), and composite adverse fetal outcomes (RR = 0.65, 95% CI 0.36–1.19) were lower in the DP group, respectively, compared with the SP group, although no level of significance was reached. Further, subgroup analyses by gravidity for low birth weight, small for the gestation age, and composite adverse fetal outcomes by gravidity resulted still in nonsignificant changes in the risk for either subgroup (Table 3).

The two study drugs were well tolerated by most participants, while the incidence of elevated ALT at 28 weeks of gestation did not differ significantly between both groups (3.8% in the SP group vs. 1.6% in the DP group, $P=0.586$). No clinical adverse events consistent with hepatotoxicity occurred during the course of the trial. On the contrary, the incidence of nausea and vomiting was significantly observed in four participants in the SP group compared to none in the DP group (4.3% vs. 0.0%, $P=0.041$) (Table 4).

Discussion

To our knowledge, this is the first clinical trial that evaluated IPTp with DP as an alternative to IPTp with SP in West Africa, including Nigeria. In the present trial, there was no significant difference in the risk of

Table 2 Intention-to-treat analysis results of the primary and secondary outcomes ($N = 206$)

Outcomes	Gravidity	Trial groups		Crude RR (95% CI)[†]	Adjusted RR (95% CI)[†]
		SP ($n = 94$) *n* (%)	DP ($n = 112$) *n* (%)		
Mean gestational age at delivery (weeks)	All gravidae	38.7 ± 2.4	38.85 ± 2.46	–	–
	Primigravidae	38.6 ± 3.5	39.3 ± 1.4	–	–
	Multigravidae	38.8 ± 1.6	38.6 ± 2.8	–	–
Rogerson placental malaria category					
None	All gravidae	46 (48.9)	41 (36.9)	1.30 (0.97–1.75)	-
Acute	All gravidae	0 (0.0)	6 (5.4)	-	-
Chronic	All gravidae	20 (21.3)	27 (24.3)	1.10 (0.76–1.60)	-
Past	All gravidae	28 (29.8)	37 (33.4)	1.09 (0.79–1.52)	-
Placental malaria[a]	All gravidae	48 (51.1)	70 (62.5)	1.29 (0.96–1.73)	1.27 (0.94–1.71)
	Primigravidae	13 (39.4)	27 (71.1)	1.99 (1.18–3.33)*	1.89 (1.13–3.16)*
	Multigravidae	35 (57.4)	43 (58.9)	1.02(0.70–1.48)	0.97 (0.67–1.41)
Spontaneous abortion	All gravidae	2 (2.1)	1 (0.9)	0.68 (0.30–1.54)	-
	Primigravidae	2 (6.1)	0 (0.0)	0.45 (0.35–0.58)*	-
	Multigravidae	0 (0.0)	1 (1.4)	-	-
Stillbirth	All gravidae	0 (0.0)	0 (0.0)	-	-
	Primigravidae	0 (0.0)	0 (0.0)	-	-
	Multigravidae	0 (0.0)	0 (0.0)	-	-
Low birth weight[b]	All gravidae	15 (16.0)	11 (9.8)	0.76 (0.53–1.10)	0.75 (0.51–1.09)
	Primigravidae	5 (15.2)	2 (5.3)	0.61 (0.36–1.06)	-
	Multigravidae	10 (16.4)	9 (12.2)	0.84 (0.52–1.34)	0.62 (0.34–1.14)
Preterm delivery	All gravidae	5 (5.4)	6 (5.4)	1.01 (0.52–1.95)	-
	Primigravidae	0 (0.0)	0 (0.0)	-	-
	Multigravidae	5 (8.2)	6 (8.2)	0.99 (0.51–1.95)	-
Small for gestational age[b]	All gravidae	3 (3.3)	4 (3.6)	1.07 (0.45–2.54)	1.12 (0.46–2.71)
	Primigravidae	0 (0.0)	1 (2.6)	-	-
	Multigravidae	3 (4.9)	3 (4.1)	0.90 (0.40–2.05)	0.98 (0.42–2.29)
Congenital malformation	All gravidae	0 (0.0)	0 (0.0)	-	-
	Primigravidae	0 (0.0)	0 (0.0)	-	-
	Multigravidae	0 (0.0)	0 (0.0)	-	-
Low APGAR score	All gravidae	5 (5.3)	0 (0.0)	0.45 (0.38–0.52)*	-
	Primigravidae	2 (6.1)	0 (0.0)	0.45 (0.35–0.58)*	-
	Multigravidae	3 (4.9)	0 (0.0)	0.44 (0.37–0.54)*	-
[#]Composite adverse birth outcome[b]	All gravidae	15 (16.0)	13 (11.6)	0.83 (0.57–1.21)	0.82 (0.55–1.21)
	Primigravidae	5 (15.2)	2 (5.3)	0.61 (0.36–1.06)	-
	Multigravidae	10 (16.4)	11 (14.9)	0.94 (0.57–1.54)	0.97 (0.59–1.61)

APGAR Appearance, Pulse, Grimace, Activity, and Respiration

[a] Adjusted for gravidity, baseline maternal anemia, and age-group

[b] Adjusted for gravidity, baseline maternal anemia, age-group and BMI category

[#] Any adverse birth outcome (spontaneous abortion, low birth weight, preterm delivery, and small for gestational age) is included

[†] SP group is the reference group for Relative Risk (RR) calculations

*Significant at $P < 0.05$

placental malaria between DP and SP. Conversely, based on point estimates DP was associated with a significant lower risk of a low APGAR score. Also, nonsignificant lower risks of low birth weight, preterm delivery, small for the gestational age, and a composite of any adverse birth outcome were noted in the DP group. The adverse effects of both study drugs were similar, although a

Table 3 Per protocol analysis results of the primary and secondary outcomes ($N=84$)

Outcomes	Gravidity	Trial groups		Crude RR (95% CI)[†]
		SP ($n=36$) n (%)	DP ($n=48$) n (%)	
Mean gestational age at delivery (weeks)	All gravidae	39.06 ± 1.59	39.63 ± 1.30	–
	Primigravidae	39.36 ± 0.81	39.67 ± 1.09	–
	Multigravidae	38.92 ± 1.82	39.60 ± 1.43	–
Rogerson placental malaria category				
None	All gravidae	17 (47.2)	21 (43.8)	1.08 (0.66–1.77)
Acute	All gravidae	0 (0.0)	4 (8.3)	–
Chronic	All gravidae	5 (13.9)	13 (27.1)	1.69 (0.77–3.72)
Past	All gravidae	14 (38.9)	10 (20.8)	0.63 (0.39–1.01)
Placental malaria	All gravidae	19 (52.8)	27 (56.2)	1.08 (0.66–1.77)
	Primigravidae	5 (45.5)	10 (55.6)	1.29 (0.50–3.28)
	Multigravidae	14 (56.0)	17 (56.7)	1.02 (0.57–1.82)
Stillbirth	All gravidae	0 (0.0)	0 (0.0)	–
	Primigravidae	0 (0.0)	0 (0.0)	–
	Multigravidae	0 (0.0)	0 (0.0)	–
Low birth weight	All gravidae	5 (13.9)	3 (6.2)	0.65 (0.36–1.19)
	Primigravidae	0 (0.0)	1 (5.6)	–
	Multigravidae	5 (20.0)	2 (6.7)	0.58 (0.33–1.04)
Preterm delivery	All gravidae	2 (5.6)	1 (2.1)	0.63 (0.27–1.46)
	Primigravidae	0 (0.0)	0 (0.0)	–
	Multigravidae	2 (8.0)	1 (3.3)	0.66 (0.28–1.56)
Small for gestational age	All gravidae	2 (5.6)	1 (2.1)	0.63 (0.27–1.46)
	Primigravidae	0 (0.0)	0 (0.0)	–
	Multigravidae	2 (8.0)	1 (3.3)	0.66 (0.28–1.56)
Congenital malformation	All gravidae	0 (0.0)	0 (0.0)	–
	Primigravidae	0 (0.0)	0 (0.0)	–
	Multigravidae	0 (0.0)	0 (0.0)	–
Low APGAR score	All gravidae	2 (5.6)	0 (0.0)	0.42 (0.32–0.52)
	Primigravidae	0 (0.0)	0 (0.0)	–
	Multigravidae	2 (8.0)	0 (0.0)	0.43 (0.32–0.59)
Composite adverse birth outcome[#]	All gravidae	5 (13.9)	3 (6.2)	0.65 (0.36–1.19)
	Primigravidae	0 (0.0)	1 (5.6)	–
	Multigravidae	5 (20.0)	2 (6.7)	0.58 (0.33–1.04)

APGAR Appearance, Pulse, Grimace, Activity, and Respiration

[#] Any adverse birth outcome (low birth weight, preterm delivery, congenital anomaly, and small for gestational age) is included

[†] SP group is the reference group for Relative Risk (RR) calculations

significant occurrence of nausea and vomiting was observed in the SP group.

In the present trial, there was no significant difference in the risk of placental malaria between both study groups, although there was a slightly increased risk of placental malaria in the DP group compared with the SP group. Relatively low SP resistance in West African countries including Nigeria and longer intervals between courses of DP due to its less frequent dosing could be responsible for this finding. This suggests that DP may likely have provided partial protection against malaria possibly by suppressing parasite densities in the placenta rather than clearing them. This finding is not in agreement with the findings in the three-dose DP arm of previous Tanzanian and Ugandan trials [9, 10]. Differences in the SP resistance levels across African countries and study designs could be responsible for the different results. In this trial, DP was associated with a significant lower risk of a low APGAR score. To our knowledge, the present study is the first to report this finding. This finding could be due to mechanisms not mediated by malaria. Hence, further studies are recommended to unravel this

Table 4 Adverse effects of the study drugs

Adverse effects	Total *N* (%)	SP *n* (%)	DP *n* (%)	*P* value
Elevated maternal ALT level	3/116 (2.6)	2/52 (3.8)	1/64 (1.6)	0.586[a]
Fever	9/203 (4.4)	4/92 (4.3)	5/111 (4.5)	1.000[a]
Fatigue	1/203 (0.5)	0/92 (0.0)	1/111 (0.9)	1.000[a]
Headache	3/203 (1.5)	0/92 (0.0)	3/111 (2.7)	0.253[a]
Skin rash	2/203 (1.0)	0/92 (0.0)	2/111 (1.8)	0.502[a]
Nausea/Vomiting	4/203 (2.0)	4/92 (4.3)	0/111 (0.0)	0.041*[a]
Dizziness	2/203 (1.0)	2/92 (2.2)	0/111 (0.0)	0.204[a]

ALT Alanine aminotransferase

[a] Fisher's exact test

*Significant at $P < 0.05$

mechanism. Also, the present trial showed a lower incidence of low birth weight in the DP group relative to the SP group. Despite the similar gestational age at birth, this finding suggests that the higher birth weight noted could also be due to mechanisms not mediated by malaria. Urogenital schistosomiasis is common in the study area [17]. Evidence has shown that intestinal schistosomiasis is associated with adverse birth outcomes [18, 19]. DP has been found to be effective against intestinal schistosomiasis [20]. This may partly explain the observed non-malaria effect of DP, although the exact non-malarial mechanisms by which DP is improving birth weight are still not clear and warrant further study. However, the result of the present trial is in agreement with that of a more recent superiority trial of monthly IPTp with DP versus monthly IPTp with SP in Tanzania and Uganda [9, 11]. In contrast, results of other previous trials in Uganda and Kenya showed higher risks of low birth weight among neonates of women in intermittent screening and treatment with DP group, and a three-dose or monthly IPTp with DP groups [10, 12]. Moreover, a pooled analysis of individual participant-level data from these three trials conducted in Kenya and Uganda showed that SP conferred a greater non-malarial effect on birth weight than DP [21]. Differences in the trial designs and participants-specific risk factors could account for these observed differences.

Overall, the present trial showed no significantly lower risk of a composite of any adverse birth outcomes in the DP group compared with the SP group consistent with a similar study conducted in Tanzania [9]. This finding could also be due to mechanisms not mediated by malaria. On the contrary, three previous randomized trials showed higher incidences of composite adverse birth outcomes in the DP group than in the SP group [10–12]. Safety and side effects are important considerations when preventive drugs are being evaluated for routine use during pregnancy. In the present trial, the overall incidence of adverse drug events was comparable in the two trial arms except for nausea and vomiting. The incidence of nausea and vomiting was significant in the SP group. This finding is comparable to nausea and stomach upset reported previously [22]. More so, available data have shown that SP can lead to a transient rise of liver enzymes [23]. On the other hand, despite piperaquine having long been associated with QTc prolongation [24, 25], evidence shows that it has the capacity to cause elevation of liver enzymes, especially ALT [26]. In the present trial, there was no significant difference in the risk of elevation of ALT between the two treatment groups. Although, a higher elevation and more participants with values higher than the upper normal limit were noted in the SP group compared to the DP group with no clinical or statistical differences. In contrast, a previous trial did not report similar findings [11].

Limitations

The main limitation of the present trial is the small sample size, low adherence to all three courses of IPTp, and lack of data on drug resistance in the cohort. Also, the exclusion of RDT-positive women may limit the generalizability of the findings. Another limitation is that the trial was not powered to detect differences in birth outcomes. Furthermore, DP has been shown to cause prolongation of the QTc interval; however, the QTc interval was not evaluated in this trial. Another limitation is that IPTp is supposed to be directly observed therapy but due to multiple doses required per course of the trial drugs, participants were adequately instructed to take the drugs at home. Therefore, 100% adherence to the trial drugs may not have been achieved.

Conclusions

In conclusion, IPTp with DP was safe and well tolerated. The risk of placental malaria was not significantly different between IPTp with DP and IPTp with SP. Also, the risks of adverse birth outcomes were not significantly different between both study drugs except for a low APGAR score. These findings add to a growing body of literature indicating that DP is a promising alternative to SP for IPTp in areas with a high level of malaria parasite resistance to SP. In spite of this contribution to knowledge, future trials are recommended to confirm the findings of the present trial.

Abbreviations

ACT Artemisinin-based combination therapy
ALT Alanine aminotransferase
APGAR Appearance, pulse, grimace, activity, and respiration
dhfr Dihydrofolate reductase
dhps Dihydropteroate synthase

DOT	Directly observed therapy
DP	Dihydroartemisinin–piperaquine
HIV	Human immunodeficiency virus
IPTp	Intermittent preventive treatment in pregnancy
ITT	Intention-to-treat
PP	Per protocol
RDT	Rapid diagnostic test
RR	Relative risk
SP	Sulfadoxine–pyrimethamine

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Author contributions
RNO conceived and designed the trial, and also acquired funding. JDO and TSY conducted literature review and participated in writing the proposal. ADG acquired the clinical data, conducted participants' follow-up and supervised the trial. AAB analyzed the blood specimens and interpreted the hematological results. ABZ and ABM performed the histological examination of the placental tissues and interpreted the results. RNO prepared a draft of the manuscript, edited and prepared the final copy of the manuscript. All authors read and approved the final manuscript.

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Availability of data and materials
All data and materials are available from the corresponding author upon request.

Declarations

Ethics approval and consent to participate
All participants provided written informed consent voluntarily. The trial was approved by the Health Research Ethics Committee of the University of Maiduguri Teaching Hospital, Maiduguri, Nigeria (UMTH/REC/555). Trial Registration: This study is registered at https://pactr.samrc.ac.za (The Pan African Clinical Trials Registry Identifier: PACTR202002644579177). All the authors vouch for the completeness and accuracy of the data and analyses presented and for the fidelity of the trial to the protocol.

Consent for publication
All the authors approved the manuscript for publication. Also, the participants gave consent for the publication of this manuscript.

Competing interests
The authors declare that they have no competing interests.

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References

1. Breman JG, Alilio MS, Mills A (2004) Conquering the intolerable burden of malaria: what's new, what's needed: a summary. Am J Trop Med Hyg 71(2 Suppl):1–15
2. Ramharter M, Grobusch MP, Kiessling G, Adegnika AA, Moller U, Agnandji ST et al (2005) Clinical and parasitological characteristics of puerperal malaria. J Infect Dis 191(6):1005–1009
3. World Health Organization (2004) A strategic framework for malaria prevention and control during pregnancy in the African region. WHO Regional Office for Africa, Brazzaville
4. Naidoo I, Roper C (2011) Drug resistance maps to guide intermittent preventive treatment of malaria in African infants. Parasitology 138(12):1469–1479
5. Thar R, Basco LK (2006) Molecular epidemiology of malaria in Cameroon. XXII. Geographic mapping and distribution of *Plasmodium falciparum* dihydrofolate reductase (DHFR) mutant alleles. Am J Trop Med Hyg 75(3):396–401
6. Oguike MC, Falade CO, Shu E, Enato IG, Watila I, Baba ES et al (2016) Molecular determinants of sulfadoxine–pyrimethamine resistance in *Plasmodium falciparum* in Nigeria and the regional emergence of dhps 431V. Int J Parasitol Drugs Drug Resist 6(3):220–229
7. Nambozi M, Mulenga M, Halidou T, Tagbor H, Mwapasa V, Phiri LK et al (2015) Safe and efficacious artemisinin-based combination treatments for African pregnant women with malaria: a multicentre randomized control trial. Reprod Health 12:5. https://doi.org/10.1186/1742-4755-12-5
8. World Health Organization (2015) Guidelines for the treatment of malaria. World Health Organization, Geneva
9. Mlugu EM, Minzi O, Kamuhabwa AAR, Aklillu E (2021) Effectiveness of intermittent preventive treatment with dihydroartemisinin–piperaquime against malaria in pregnancy in Tanzania: a randomized controlled Trial. Clin Pharmacol Ther 110(6):1478–1489
10. Kakuru A, Jagannathan P, Muhindo MK, Natureeba P, Awori P, Nakalembe M et al (2016) Dihydroartemisinin–piperaquine for the prevention of malaria in pregnancy. N Engl J Med 374(10):928–939
11. Kajubi R, Ochieng T, Kakuru A, Jagannathan P, Nakalembe M, Ruel T et al (2019) Monthly sulfadoxine–pyrimethamine versus dihydroartemisinin–piperaquine for intermittent preventive treatment of malaria in pregnancy: a double-blind, randomised, controlled, superiority trial. Lancet 393(10179):1428–1439
12. Desai M, Gutman J, Llanziva A, Otieno K, Juma E, Kariuki S et al (2015) Intermittent screening and treatment or intermittent preventive treatment with dihydroartemisinin-piperaquine versus intermittent preventive treatment with sulfadoxine-pyrimethamine for the control of malaria during pregnancy in western Kenya: an open-label, three-group, randomised controlled superiority trial. Lancet 386(10012):2507–2519
13. Nigeria National Malaria Elimination Programme (2018) LSHTM and KEMRI-Wellcome Trust Research Programme. Nigeria: a Profile of malaria control and epidemiology, 2018 overview. A report prepared for the Federal Ministry of Health, Nigeria and the Department for International Developments, UK
14. Balogun ST, Sandabe UK, Sodipo OA, Okon KO, Akanmu AO (2021) Single nucleotide polymorphisms of Pfdhfr and Pfdhps genes: implications for malaria prophylactic strategies in Maiduguri, Northeast Nigeria. J Trop Med. https://doi.org/10.1155/2021/8840089
15. Natureeba P, Ades V, Luwedde F, Mwesigwa J, Plenty A, Okong P et al (2014) Lopinavir/ritonavir-based antiretroviral treatment (ART) versus efavirenz-based ART for the prevention of malaria among HIV-infected pregnant women. J Infect Dis 210(12):1938–1945
16. Rogerson SJ, Hviid L, Duffy PE, Leke RFG, Taylor DW (2007) Malaria in pregnancy: pathogenesis and immunity. Lancet Infect Dis 7(2):105–117
17. Babagana U, Pukuma MS (2018) Epidemiology of schistosomiasis in Damboa, Gamboru and Baga IDP camps in Maiduguri. Borno State Int J Res Publ 13(1):12
18. Friedman JF, Mital P, Kanzaria HK, Olds GR, Kurtis JD (2007) Schistosomiasis and pregnancy. Trends Parasitol 23(4):159–164
19. Mombo-Ngoma G, Honkpehedji J, Basra A, Mackanga JR, Zoleko RM, Zinsou J et al (2017) Urogenital schistosomiasis during pregnancy is associated with low birth weight delivery: analysis of a prospective cohort of pregnant women and their offspring in Gabon. Int J Parasitol 47(1):69–74
20. Mnkugwe RH, Minzi O, Kinung'hi S, Kamuhabwa A, Aklilu E (2020) Efficacy and safety of praziquantel and dihydroartemisinin-piperaquine

combination for treatment and control of intestinal schistosomiasis: a randomized, non-inferiority clinical trial. PLoS Negl Trop Dis 14(9):e0008619
21. Roh ME, ter Kuile FO, Rerolle F, Glymour MM, Shiboski S, Gosling R et al (2020) Overall, anti-malarial, and non-malarial effect of intermittent preventive treatment during pregnancy with sulfadoxine–pyrimethamine on birthweight: a mediation analysis. Lancet Glob Health 8(7):e942–e953
22. Maiga As Diakite M, Diawara A, Sango HA, Coulibaly CO (2010) Pharmacovigilance and impact of intermittent preventive treatment with sulfadoxine–pyrimethamine in pregnant women in Selingue. Mali Mali Med 25(3):41–48
23. RxList. Fansidar (2017) https://www.rxlist.com/fansida-drug.htm. Accessed 25 Jul 2022
24. White NJ (2007) Cardiotoxicity of antimalarial drugs. Lancet Infect Dis 7(8):549–558
25. World Health Organization (2017) Malaria policy advisory committee meeting: the cardiotoxicity of antimalarials. http://www.who.int/malaria/mpac/mpacmar2017-erg-cardiotoxicity-report-session2.pdf. Accessed 25 May 2022.
26. Chughlay MF, Akakpo S, Odedra A, Csermak-Renner K, Djeriou E, Winnips C et al (2020) Liver enzyme elevations in plasmodium falciparum volunteer infection studies: findings and recommendations. Am J Trop Med Hyg 103(1):378–393

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