

Research Article

Per protocol analysis of therapeutic efficacy of chloroquine for the treatment of *Plasmodium vivax* malaria in children

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ABSTRACT

Background: High incidence of chloroquine-resistant vivax malaria has led to increasing case fatality and incidence of complicated malaria with *Plasmodium vivax* contributing to morbidity and mortality. Current guidelines recommend the use of chloroquine along with tissue schizonticides like primaquine for the treatment of *Plasmodium vivax* malaria. There were reports of increasing resistance to chloroquine in *Plasmodium vivax* species especially from Asia during the period when this study was taken up.

Methods: Present study is a one arm, 28 day follow up, hospital based prospective observational study including 150 children aged 1 to 12 years with smear positive isolated *Plasmodium vivax* infections. Chloroquine was administered at the standard dose of 25 mg base/kg body weight over three days. Recurrence of parasitaemia and clinical conditions of patients were assessed on days 1, 2, 3, 7, 14, and 28 during the 28-day follow-up period.

Results: All 150 children included in the study, completed their 28 day follow up. Mean age for enrolment was 5.96 ± 2.38 years. The mean parasite index (%) on enrolment was 1.22 ± 0.46 . Based on the per protocol analysis, chloroquine efficacy (adequate clinical and parasitological response) was 92.7%. Treatment was well tolerated. Early treatment failure was seen in 2.7% of study population, late clinical failure in 2% and late parasitological failure in 2.7% of study population, suggesting that response to chloroquine was good in the study population.

Conclusions: In the present study the chloroquine therapy showed high therapeutic efficacy (92.7%) in the treatment of uncomplicated vivax malaria. It was also well tolerated and equally safe when used in therapeutic doses. Regular monitoring of the pattern of resistance to chloroquine is needed in vivax malaria endemic areas of the country and measures are taken rapidly and effectively to control spread of resistance.

Keywords: *Plasmodium vivax*, Chloroquine resistance, Therapeutic efficacy, Treatment failure

INTRODUCTION

Plasmodium vivax is most widespread and results in pronounced morbidity. India is a major contributor to the burden of vivax malaria.¹ Till recently *P. vivax* has remained uniformly sensitive to chloroquine, and this cheap and widely available antimalarial drug has been the treatment of choice for the past 50 years. Since the recent studies have suggested both ability of *P. vivax* to cause a

severe disease and the emergence of resistance to chloroquine in these species, it is of utmost importance to know the degree of response of the parasite to this drug.²⁻⁴ There are very few Indian studies which mention about the burden of chloroquine resistance in *P. vivax* malaria and none in the pediatric age group. Therefore the present study has been designated to assess the current status of clinical and parasitological response to chloroquine in vivax malaria in children.

METHODS

This study was designed as a prospective observational study, conducted over 22 months from January 2013 to October 2014 at a major tertiary care teaching hospital, primarily catering to the urban-slum population of a cosmopolitan city. The study included smear-positive cases of isolated *Plasmodium vivax* infection admitted in the pediatric ward.

A sample size of 150 cases of uncomplicated vivax malaria was considered as adequate for this study, presuming an expected proportion of chloroquine resistance in *vivax* infections as 10% ($p=0.1$) and considering a precision value (d) as 0.05, a sample size of 136 was found to be adequate, using Daniel formula: ($n=Z^2P(1-P)/d$), with Z as statistic for a level of confidence=1.84.⁵

The study was approved by ethics committee of the institute. Inclusion criteria included age ≥ 1 and <12 years, peripheral smear positive for *P.vivax* mono infection, parasite density $\geq 250/\mu\text{l}$, history of fever during 48 hours prior to time of recruitment. Cases with co-infection with *Plasmodium falciparum*, any other significant concomitant illness, and intolerance to oral chloroquine were excluded.

Fever was defined as documented axillary temperature of $\geq 37.5^\circ\text{C}$ or rectal temperature of $\geq 38^\circ\text{C}$.⁶ The parasite identification was performed using the thick and thin smears, stained with wright's stain, and the parasite index was given as parasite count per 100 red blood cells. Peripheral blood smears were collected at 12-hourly intervals till three consecutive smears were negative for malaria parasite. The enrolled patients meeting the inclusion criteria were then started on PO chloroquine with an initial dose of 10mg/kg followed by 5 mg/kg at 6, 24 and 48 hours. All children were kept in hospital till minimum D3 of therapy or at least 48 hours after defervescence and parasitological clearance. After clinical and parasitological clearance, subsequently follow up was done on outpatient basis. All patients were given per oral primaquine 0.25 mg/kg OD for 14 days. All cases were followed up as per the WHO protocol shown in Table 1, or more frequently if required. As a deviation from WHO protocol, day 21 follow up was excluded due to logistic reasons and chloroquine/metabolic drug levels or molecular studies were not done due to lack of facilities.

At the end of the study, all cases were classified for following treatment responders or treatment failure. A treatment failure was defined as follows:⁷

- Clinical deterioration in presence of parasitemia during/after hospitalization period.
- Presence of parasitemia and axillary temperature $>37.5^\circ\text{C}$ on any day between D3-28.

- Presence of parasitemia on any day between D3-28, irrespective of clinical condition.

Following secondary outcome parameters were also recorded to assess the pattern of therapeutic responses:⁷

- Fever clearance time (FCT in hours).
- Parasite clearance time (PCT in hours).
- Any significant adverse event necessitating discontinuation/change of therapy.

Statistical analysis was done using the SSPS software version of 2015. Primary and secondary outcome measures were analysed according to various study determinants using Chi-square test or Fischer test for the non-parametric data and student's test for parametric data. In all analysis, p -value <0.05 was considered significant.

Table 1: Follow-up protocol (methods).

Days	0	1	2	3	7	14	28
Clinical examination (fever)	X	X	X	X	X	X	X
Treatment with chloroquine	X	X	X				
Parasitemia	X	X	X	X	X	X	X
Hemoglobin	X						X

RESULTS

A total of 150 patients fulfilled the inclusion criteria and all of them completed the 28 day follow-up. Among the recruited study participants, males (58%) were higher in proportion compared to females (42%).

Table 2: Characteristics of *P. vivax*-infected patients enrolled in the *in vivo* therapeutic efficacy study of CQ for *plasmodium-vivax* malaria at a tertiary care centre in Mumbai, India from January 2014 to October 2015.

Characteristics	Total number
Number of enrolled patients	150
Mean age (\pm SD, range), years	5.96 \pm 2.38 (1-12 years)
Sex ratio (M/F)	1.38 (87/63)
Mean weight (\pm SD, range), kg	16.24 \pm 4.87 (7-31 kg)
Mean haemoglobin (\pm SD, range), gm/dl	9.11 \pm 1.47 (5.6-12.4 gm/dl)
Mean platelet count (\pm SD, range), lakhs/mm ³	2.09 \pm 0.74 (0.78-4.78 lakhs/mm ³)
Mean duration of fever (\pm SD, range), days	4.61 \pm 1.66 (2-10 days)
Mean parasitic index (\pm SD, range)%	1.22 \pm 0.46 (1-3%)
Mean fever clearance time (\pm SD, range), hours	19.89 \pm 7.92 (8-72 hours)
Mean parasite clearance time (\pm SD, range) hours	25.64 \pm 10.79 (12-72 hours)

The median age and weight of study participants was 6 years (range 1 to 12 years) and 16 kg (range of 7 to 31 kg) respectively (Table 2).

Out of 150 patients, 4 patients suffered from adverse drug reaction following chloroquine ingestion. 2 of them had nausea, 1 had vomiting and 1 had myalgia, however that didn't require to stop the chloroquine and they all had adequate clinical response to chloroquine. The mean duration of fever amongst the study population was 4.61 ± 1.66 (mean \pm standard deviation (SD)) days. Mean haemoglobin level of the study population was 9.11 ± 1.47 (mean \pm SD) gm/dl. Mean parasite index of the study population was 1.22 ± 0.46 (mean \pm SD)%. All study participants cleared fever following parasitemia clearance (Table 2). Mean fever clearance time and parasite clearance time for the study population was 19.89 ± 7.92 hours and 25.64 ± 10.79 hours respectively. Adequate clinico-parasitological response (ACPR) was achieved in 92.7% (N=139) of children receiving chloroquine. Early treatment failure (ETF) was seen in 2.7% (N=4) of study population, late clinical failure (LCF) in 2% (N=3) and late parasitological failure (LPF) in 2.7% (N=4) of the study population, suggesting that the response to chloroquine was good in the study population. Adequate clinical response was achieved in 58% of males and 42% of females in the study population.

Correlation between clinical features and therapeutic response of the study population

Mean age for enrolment in the present study was 5.96 ± 2.38 (mean \pm SD) years. In the present study, the mean age for ACPR was 6.04 ± 2.40 years. The mean age for late parasitological failure is 6.75 ± 2.2 years. The mean age for early treatment failure and late clinical failure was 4 ± 0.82 years and 4 ± 0.00 years respectively. The differences in these responses was however not clinically significant ($p=0.101$).

There was marginal male preponderance in showing response to chloroquine adequately (56.8% vs 43.2%) and also early treatment failure being more in males than females (75% vs 25%). Late clinical failure was seen only in males and late parasitological failure was equally distributed in both the groups. The sex-difference however was not statistically significant ($p=0.419$).

Although nutritional status could be an important confounder for drug efficacy and toxicity, it has been rarely considered as an important case variable in anti-malarial therapeutic efficacy studies except for occasional comment. Mean weight of the study population was 16.24 kg. There was no correlation between weight and duration of fever or parasitic index in the study population ($p=0.139$).

Previous history of chloroquine intake was taken into consideration while taking into account the clinical

response to chloroquine in the present illness. There was no significant difference in the clinical response as judged by ACPR, FCT, PCT with relation to previous history of chloroquine ingestion (Table 3).

Table 3: Per-protocol analysis of chloroquine efficacy to treat *P. vivax* malaria in the study population.

Outcome	Total number
Per protocol population, n	150
Lost-to-follow-up, n (%)	0
Early treatment failure (ETF), n (%)	4 (2.7%)
Late clinical failure (LCF), n (%)	3 (2.0%)
Late parasitological failure (LPF), n (%)	4 (2.7%)
Adequate clinical and parasitological response (ACPR), n (%)	139 (92.7%)

DISCUSSION

Anemia was a common finding in patients with *P. vivax* malaria. It is an important, but incompletely understood cause of anemia. Young children are at high risk of clinically significant and potentially severe *vivax*-associated anemia, particularly in countries where transmission is intense and relapses are frequent. Severe *vivax* anemia is associated with substantial indirect mortality and morbidity. Anemia can be averted by early and effective anti-malarial treatment.⁸ According to a study in children done in Africa, on clinical evaluations 94.87% cases presented with anemia, out of which 10.26% were severe, the mean haemoglobin levels on admission were 8.09 g/dl and no deaths were reported.⁹ In the study the mean hemoglobin of study population was 9.1 ± 1.47 g/dl. There was no correlation between anemia and fever clearance time or parasitic index according to this study. There was no significant correlation of ACPR, ETF, LCF and LPF with age, weight, sex, history of chloroquine intake or duration of fever, in the present study.

This study is the first evaluation of chloroquine efficacy against *P. vivax* malaria done exclusively on 150 children less than 12 years in India. The results indicate that chloroquine is highly efficacious and well tolerated in the *vivax* endemic region of the study.^{10,11} Although chloroquine was highly effective as per our study, chloroquine resistant *P. vivax* has been reported from foci in South east Asia, India, Thailand, Egypt, South America and more recently from Madagascar and Ethiopia in Eastern Africa.¹²⁻¹⁹ Chloroquine is being used for treatment as well as prophylaxis in health sector in India. It is the anti-malarial drug recommended as the first line treatment for *vivax* malaria by the National Drug Policy of malaria in India.¹³ However there are some threatening reports on chloroquine resistant *vivax* malaria from different malaria endemic regions of India.¹⁴⁻³⁰ Thus it is becoming a major public health problem that needs to be dealt with on an urgent basis by initiating proper

diagnosis of cases and administration of proper dosage and duration of effective anti-malarials.¹⁵

Fever clearance time in the study population

Fever clearance time is the time between onset of treatment and return of temperature to normal. It is taken as one of the variable to assess the therapeutic efficacy of antimalarial drugs though it is a crude method. Since early treatment failure is defined as persistence of fever or parasitemia for more than seven days, fever clearance time is an important variable to assess the efficacy of chloroquine in treatment of *Plasmodium vivax* malaria. In the present study fever clearance time was statistically significant in cases of treatment failure and ACPR varied as the fever clearance time increased. In cases of early treatment failure the mean fever clearance time was 42 hours. It was statistically significant. Also prolonged fever clearance time was significantly associated with a higher parasitic index. Whereas in studies from Thailand cure rate with chloroquine was 100%, no recrudescence was observed during the 28 day follow up.^{10,11} There were no significant correlations with parasitic index and fever clearance time.

The study shows 11 cases of *vivax* malaria out of total 150 study population as chloroquine treatment failures (7.3%). It is important to alert the local government bodies of all the malaria endemic region of the country to monitor drug resistance and address treatment failure problems. According to WHO, first-line treatment of malaria should be changed if the total failure rate is more than 10%.¹⁵ It is important to analyse CQ treatment failures and intervene before the level is attained that presents a major public health problem.

CONCLUSION

Thus the present study supports the current guidelines for the management of *vivax* malaria in children, recommending chloroquine as the first drug of choice with tissue schizonticides like primaquine given for 14 days. Hence the above therapy should be used in clinical practice as per the WHO/Government of India guidelines to avoid emergence and spread of drug resistance.

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REFERENCES

- Joshi H, Prajapati SK, Verma A, Kang'a S, Carlton JM. *Plasmodium vivax* in India. Trends Parasitol. 2008;24(5):228-35.
- Pukrittayakamee S, Chantira A, Simpson JA, Vanijanonta S, Clemens R, Looareesuwan S, et al. Therapeutic responses to different antimalarial drugs in *vivax* malaria. Antimicrob Agents Chemother. 2000;44(6):1680-5.
- Manju K, Radha G, Kumari M, Ghildiyal R. Clinical profile of *Plasmodium vivax* malaria in children and study of severity parameters in relation to mortality: a tertiary care centre perspective in Mumbai, India. Malar Res Treat. 2014;2014:765657.
- Price RN, Seidlein L, Valecha N, Nosten F, Baird JK, White NJ. Global extent of chloroquine-resistant *Plasmodium vivax*: a systematic review and meta-analysis. Lancet Infect Dis. 2014;14(10):982-91.
- Indrayan A, Satyanarayana L. Basic philosophy of statistical tests, confidence intervals and sample size determination. Indian Pediatr. 2000;37(7):739-51.
- World Health Organization, WHO guidelines for the treatment of malaria. Geneva, World Health Organisation;2010.
- World Health Organization. Monitoring Antimalarial Drug Resistance, Report of a WHO consultation. WHO/CDS/CSR/EPH/2002. Geneva, World Health Organisation;2002.
- Douglas NM, Anstey NM, Buffet PA, Poespoprodjo JR, Yeo TW, White NJ, et al. The anaemia of *Plasmodium vivax* malaria. Malar J. 2012;11:135.
- Rodríguez MAJ, Sánchez E, Vargas M, Piccolo C, Colina R, Arria M. Anemia and thrombocytopenia in children with *Plasmodium vivax* malaria. J Trop Pediatr. 2006;52(1):49-51.
- Congpuong K, Bangchang NK, Thimasarn K, Tasanor U, Wernsdorfer WH. Sensitivity of *Plasmodium vivax* to chloroquine in Sa Kaeo Province, Thailand. Acta Trop. 2002;83(2):117-21.
- Tasanor O, Ruengweeraut R, Sirichaisinthop J, Congpuong K, Wernsdorfer WH, Bangchang NK. Clinical-parasitological response and *in-vitro* sensitivity of *Plasmodium vivax* to chloroquine and quinine on the western border of Thailand. Trans R Soc Trop Med Hyg. 2006;100(5):410-8.
- Baird JK. Neglect of *Plasmodium vivax* malaria. Trends Parasitol. 2007;23(11):533-9.
- Barnadas C, Ratsimbaoa A, Tichit M, Bouchier C, Jahevitra M, Picot S, et al. *Plasmodium vivax* resistance to chloroquine in Madagascar: clinical efficacy and polymorphisms in *pvmr1* and *pvcrt-o* genes. Antimicrob Agents Chemother. 2008;52(12):4233-40.
- Teka H, Petros B, Yamuah L, Tesfaye G, Elhassan I, Muchohi S, et al. Chloroquine-resistant *Plasmodium vivax* malaria in Debre Zeit, Ethiopia. Malar J. 2008;7:220.
- Yohannes AM, Teklehaimanot A, Bergqvist Y, Ringwald P. Confirmed *vivax* resistance to chloroquine and effectiveness of artemether-lumefantrine for the treatment of *vivax* malaria in Ethiopia. Am J Trop Med Hyg. 2011;84(1):137-40.
- Hwang J, Alemayehu BH, Reithinger R, Tekleyohannes SG, Takele T, Birhanu SG, et al. *In vivo* efficacy of artemether-lumefantrine and chloroquine against *Plasmodium vivax*: a

- randomized open label trial in central Ethiopia. PLoS One. 2013;8(5):e63433.
17. Assefa M, Eshetu T, Biruksew A. Therapeutic efficacy of chloroquine for the treatment of *Plasmodium vivax* malaria among outpatients at Hossana health care centre, southern Ethiopia. Malar J. BioMed Central. 2015;14(1):458.
 18. Gonsalves LA, Cravo P, Ferreira MU. Emerging *Plasmodium vivax* resistance to chloroquine in South America: an overview. Mem Inst Oswaldo Cruz. 2014;109(5):534-9.
 19. Salem OA, Lemine MYO, Deida JM, Lemrabott MA, Ouldabdallahi M, Ba MD, et al. Efficacy of chloroquine for the treatment of *Plasmodium vivax* in the Saharan zone in Mauritania. Malar J. 2015;14(1):39.
 20. Government of India. Ministry of Health and Family welfare, NVBDCP, National Drug Policy on Malaria. Delhi; 2010.
 21. Dua VK, Kar PK, Sharma VP. Chloroquine resistant *Plasmodium vivax* malaria in India. Trop Med Int Health. 1996;1(6):816-9.
 22. Mohan K, Maithani MM. Congenital malaria due to chloroquine-resistant *Plasmodium vivax*: a case report. J Trop Pediatr. 2010;56(6):454-5.
 23. Kshirsagar NA, Gogtay NJ, Rajgor D, Dalvi SS, Wakde M. An unusual case of multidrug-resistant *Plasmodium vivax* malaria in Mumbai (Bombay), India. Ann Trop Med Parasitol. 2000;94(2):189-90.
 24. Zaki SA. Chloroquine resistant *vivax* malaria in an infant: a report from India. J Vector Borne Dis. 2009;46(1):83.
 25. Singh RK. Emergence of chloroquine-resistant *vivax* malaria in south Bihar (India). Trans R Soc Trop Med Hyg. 2016;94(3):327.
 26. Shah I. Chloroquine resistant *vivax* malaria in an infant: A report from India. J Vector Borne Dis. 2008;46(2):168-9.
 27. Garg M, Gopinathan N, Bodhe P, Kshirsagar NA. *Vivax* malaria resistant to chloroquine: case reports from Bombay. Trans R Soc Trop Med Hyg. 1995;89(6):656-7.
 28. Srivastava HC, Yadav RS, Joshi H, Valecha N, Mallick PK, Prajapati SK, et al. Therapeutic responses of *Plasmodium vivax* and *P. falciparum* to chloroquine, in an area of western India where *P. vivax* predominates. Ann Trop Med Parasitol. 2008;102(6):471-80.
 29. Ganguly S, Saha P, Guha SK, Das S, Bera DK, Biswas A, et al. *In vivo* therapeutic efficacy of chloroquine alone or in combination with primaquine against *vivax* malaria in Kolkata, West Bengal, India, and polymorphism in PVMDR1 and PVCRT-o genes. Antimicrob Agents Chemother. 2013;57(3):1246-51.
 30. Shalini S, Chaudhuri S, Sutton PL, Mishra N, Srivastava N, David JK, et al. Chloroquine efficacy studies confirm drug susceptibility of *Plasmodium vivax* in Chennai, India. Malar J. 2014;13(1):129.

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