ACUTE MALARIA CAUSED BY MONO- AND CO-INFECTION OF PLASMODIUM VIVAX AND FALCIPARUM: A COMPARATIVE OBSERVATIONAL STUDY OF 110 CASES

Medicine

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ABSTRACT

Introduction: Malaria has been a major health problem of all tropical countries, including India. It has a wide spectrum of clinical signs, symptoms and complications.

Objective: This study was undertaken to study various clinical symptoms, signs and complications of malaria and also to compare the specie-specific complications of malaria.

Material & Methods: This cross sectional observational study was done at a tertiary care hospital in the state of Uttarakhand. It included all cases of acute malaria aged 18 years or more, hospitalized over a period of one year (from April 2015-March 2016). Details regarding clinical profile, biochemical parameters, demography, complications and outcome of each patient were compiled.

Results: Out of 110 cases of acute malaria patients 97 (88%) had Plasmodium vivax infection, one (9%) patient had P. falciparum infection while only 3 (3%) cases had co-infection with both P. vivax and P. falciparum infection. Thrombocytopenia was observed in 104 (96.5 %) cases followed by anemia in 75 (68%) cases. All the hematological and biochemical parameters were equally distributed in the etiological groups of malaria. Only mean haemoglobin levels and mean total leucocytes count differed significantly between these groups. Acute kidney injury, liver dysfunction, shock and the need for intensive care and hemodialysis were comparable in the three etiological groups. Two patients succumbed to death while one patient left against medical advice.

Conclusion: This study throws light on varied complications of acute malaria and that it can affect any system of the body. This study has re-emphasized the fact that P. vivax malaria, long considered as "benign" malaria is now an important cause of complicated malaria.

KEYWORDS

acute malaria, mortality, morbidity, hepatomegaly, splenomegaly, P. Vivax, P. falciparum

INTRODUCTION

Malarial, diarrhea, HIV/AIDS, Tuberculosis, measles, hepatitis B and pneumonia account for 85% of global infections. [1] Malarial infection is caused by species of plasmodium; P. falciparum and P. vivax account for almost all cases of malaria in India. [2] Clinical manifestations of malaria can vary from mild asymptomatic infection to fatal multi-organ involvement. These clinical symptoms are dependent on multiple factors like host factors, patient factors and parasite factors, immunity and age of patients and geographical distribution [3] Since 1990, the reported incidence of malaria in India has been 1.5-2.6 million cases with 666-1000 deaths per annum. [4] The National Vector Borne Disease Control Programme in India (NVBDCP) uses annual parasite incidence (API) as a measure to express malaria cases per thousand population. As per NVBDCP, 2004 most of the Indian states recorded API <2. [5]

We report this study from the hilly state of Uttarakhand, which is a hypoendemic zone of malaria. [6] Kumaun region reports nine species of anopheles mosquito while in Garhwal region only three species have been reported to be dominant. [7,8] Maximum number of the cases are reported in the month of August and September. [9] Most of the previous researches have focused on malaria infection by individual species, mainly P. vivax and P. falciparum. In our study, we also did a comparative analysis describing the difference in complications associated with their mono- and co-infection.

MATERIAL & METHODS

This cross-sectional observational study was carried out at a tertiary referral center of the north Indian state of Uttarakhand over a period of one year from 2015 to 2016. Informed consent was taken from the patients and institutional ethical clearance was obtained. The research was done as per the ethical standards of the responsible committee on human experimentation and with Helsinki Declaration.

110 febrile patients aged 18 years or more with acute malaria diagnosed using rapid diagnostic kits and malarial parasite on peripheral blood smear were included in the study. Detailed clinical history and examination, demographic parameters, laboratory parameters and outcome of individual patients was compiled and tabulated. All patients were categorized as having uncomplicated or severe malaria.

Severe anemia was defined as serum haemoglobin (<5gm/dl); those with platelet counts less than 150×10^9/L were designated as having thrombocytopenia. Renal impairment (renal failure) was defined as oliguria (<400ml/24hours) along with rise in serum creatinine (>3mg/dl) and no improvement with rehydration. Respiratory involvement (respiratory distress) was defined as tachypnea (>20 minutes) along with fall in saturation of oxygen <90%. Hypoglycemia was defined as plasma glucose level at <40mg/dl. Shock was defined as the systolic blood pressure <80mmHg in adults. Hyperbilirubinemia i.e., rise in bilirubin of >3mg/dl and three-fold rise in liver transaminanses was regarded as liver dysfunction. Central nervous system involvement was considered if coma persisted > 30 mins after generalized convulsion [10].

Data Management and Statistical and Analysis

Mean and median were used as the measures of central tendency and standard deviation was used as the measure of dispersion for descriptive statistics. Tests of significance used were Pearson Chi-square (using Fisher's Exact Test) for categorical variables; and Kruskal Wallis H test for continuous variables. A p-value of < 0.05 was considered as significant. The statistical software SPSS version 19 was used for analysis.

RESULTS

P. vivax was the commonest malarial specie with 97 cases of mono-infection; falciparum co-infection was observed in 3 (2.7%) cases. Males outnumbered females constituting 60.9% of all cases studied. More than 40% cases belonged to the productive age group of 25-50 years. Sixty percent (n=66) patients hailed from rural areas and were mainly engaged in agriculture. No significant association was found between residential distribution of cases and the species of malaria. As evident in table 1, 70% patients reported fever with chills; however, all patients were febrile at presentation. Other prominent symptoms were headache and vomiting. Bleeding manifestations were seen only in 3

cases. The sites of bleeding were gums, urinary tract and nasal cavity in one case each. Nearly one-fourth of the patients of malaria had hepatomegaly and splenomegaly either alone or in combination. The various symptoms recorded at the time of hospitalization were equally distributed in the etiological groups of malaria.

Table 1: Distribution of clinical features of malaria on the basis of etiological species

Symptoms	Number of cases (%)			p-value (Pearson Chi square / Fisher Exact)
	P. vivax (n=97)	P. falciparum (n=10)	Co-infection (n=3)	
Fever	97 (100)	10 (100)	3(100)	NS
Chills	64 (66)	8 (80.0)	3 (100)	NS
Body ache	12 (12.4)	0	0	NS
Headache	59 (60.8)	8 (80.0)	3 (100)	NS
Jaundice	9 (9.3)	2 (20.0)	1(33.3)	NS
Vomiting	39 (59.8)	4 (40.0)	1 (33.3)	NS
Bleeding	1 (1.0%)	2 (20.0)	0	NS
Pain abdomen	24 (24.7)	2 (20.0)	0	NS
Loose stools	10 (10.3)	3 (30.0)	0	NS
Shortness of breath	4 (4.1)	1 (10.0)	1 (33.3)	NS
Malaise	4 (4.1)	0	1 (33.3)	NS
Cough	3 (3.6)	0	0	NS
Loss of appetite	9 (9.3)	1 (10.0)	0	NS
Pedal edema	3 (3.1)	0	0	NS
Splenomegaly	20 (20.6)	3 (30.0)	1 (33.3)	NS
Oliguria	4 (4.1)	0	0	NS
Hepatomegaly	23 (24.0)	2 (20.0)	1 (33.3)	NS
Icterus	9 (9.3)	2 (20.0)	1 (33.3)	NS
Bleeding manifestation (hematuria/ epistaxis /bleeding gums)	1 (1.00)	2 (20.0)	0	<0.05

Figures in the parenthesis indicate the column percentage

Amongst the various laboratory parameters studied (table 2), thrombocytopenia was the predominant finding seen in 104 (96.5%) cases. Anemia, predominantly normocytic

Table 2: Laboratory parameters of cases of malaria on the basis of etiological species

Parameter	P. vivax (n=97)	P. falciparum (n=10)	Co-infection (n=3)	p-value (Kruskal Wallis test)
Hemoglobin (gm/dl)	11.2 ± 2.3	9.1 ± 3.1	6.1 ± 2.6	<0.05
Total leucocyte count (x 10^3/cumm)	5.2 ± 2.8	6.4 ± 4.4	14.9 ± 7.5	NS
MCV (fl)	86.0 ± 8.2	89.4 ± 6.9	93.9 ± 17.2	NS
MCH (pg)	28.4 ± 2.8	29.9 ± 3.9	30.7 ± 5.8	NS
MCHC (%)	32.4 ± 1.9	33.3 ± 2.2	32.6 ± 0.2	NS
Platelet count (x 10^3/cumm)	46.8 ± 60.7	48.5 ± 49.4	50.1 ± 57.2	NS
ESR (mm/1st hr)	29.9 ± 21.8	25.5 ± 21.9	47 ± 46.6	NS
Total bilirubin (mg/dl)	3.5 ± 5.8	3.7 ± 3.5	6.7 ± 1.1	NS
AST (IU/L)	77.8 ± 128.5	60.8 ± 43.6	60 ± 9.8	NS
ALT (IU/L)	52.9 ± 47.9	37.2 ± 21.2	18.5 ± 6.3	NS
ALP (IU/L)	117.2 ± 81.1	81.0 ± 16.8	251.0 ± 120.8	NS
Total protein (g/dl)	5.4 ± 0.9	5.9 ± 0.7	5.5	NS
Albumin (g/dl)	2.5 ± 0.6	2.2 ± 0.4	2.2 ± 0.1	NS
BUN (mg/dl)	28.9 ± 24.9	21.6 ± 12.7	64.1 ± 2.6	NS
Creatinine (mg/dl)	1.8 ± 2.1	1.3 ± 1.0	1.1 ± 0.6	NS
Sodium (mmol/L)	136.4 ± 4.1	135.5 ± 5.8	132.4 ± 4.0	NS
Potassium (mmol/L)	4.3 ± 3.4	3.8 ± 0.4	3.5 ± 0.0	NS

normochromic, was observed in 75 (68.1%) cases. Hemoglobin was

significantly lowest in the mixed infection group and total leucocyte count was highest in this group. Hence, the groups differed significantly in mean hemoglobin levels and mean total leucocyte count. The other biochemical parameters barring serum bilirubin, AST, ALT and creatinine assessed at the time of hospitalization were equally distributed in the etiological groups of malaria. Severe anemia (hemoglobin <5 gm/dl) was observed in 2 of the 3 cases with mixed infection. Overall, 75 (68.8%) cases had anemia. Thrombocytopenia was observed in 96.1% cases with malaria; severe thrombocytopenia (<20000/cumm) was observed in 33.6% cases. However, less than 10% of these cases had overt bleeding manifestations. With treatment, 97.2% (n=107) patients improved and were subsequently discharged from the hospital. Two cases succumbed to their illness while one patient left against medical advice (table 3). No statistically significant association was found in the outcome parameters when the etiological species causing malaria were compared.

Table 3: Complications and outcomes of patients with malaria on the basis of etiological species

	Number of cases (%)				p-value (Pearson Chi square/ Fisher Exact)
	P. vivax (n=97)	P. falciparum (n=10)	Co-infection (n=3)	Total cases (n=110)	
Hemodialysis	4 (4.1)	0	0	4 (3.6)	0.757
Platelet transfusion	26 (29.8)	3 (30.0)	1 (33.3)	30 (27.2)	0.949
Blood transfusion	9 (9.2)	2 (20)	1 (33.3)	12 (10.9)	0.263
Intensive Care	2 (2.0)	0	0	2 (1.8)	0.872
Shock	14 (14.4)	1 (10)	0	15 (13.6)	0.727
Acute Kidney Injury	21 (21.6)	2 (20)	1 (33.3)	24(21.8)	0.883
Liver injury	18 (18.5)	2 (20)	2 (66.7)	22 (20)	0.126
Expired/LAMA	2 (2.0)	0	1 (33.3)	3 (2.7)	0.357

Figures in the parenthesis indicate the column percentage

DISCUSSION

We studied 110 patients of malaria diagnosed by peripheral blood smear and rapid diagnostic tests of which 88.1 % cases were caused by P. vivax mono-infection and 2.7% by its co-infection with P. falciparum. In India, P. vivax accounts for 70 % of infection followed by P. falciparum (25-30 %); their co-infection accounts for the remaining 4-5% cases. [10,11]

In our study, mean age of the patient was 39.3±17.2 years and majority of the patients (43.6 %) were in the age group of 25-50 years with male predominance. Jadhav et al in their study reported a similar mean age of 37.4 ± 14.2 years. [12] We also had male predominance in our study. Male dominance in most of the studies could be attributed to the fact that males are more mobile and scantily dressed in the rural areas and hence, more exposed to mosquito bites and malaria transmission especially in Asian countries. The above-mentioned rationale is applicable to the fact that malaria is more prevalent in rural areas traditionally. Two-third of the cases studied belonged to rural areas indulging mainly in farming and open defecation due to lack of toilets at home. A similar study from South India also reported 60% cases of malaria from rural background. [13]

Fever was the cardinal and the predominant symptom at the time of presentation in malaria as has been highlighted in earlier studies [14]. The mean duration of fever in our study was 6.2 ± 3.5 days concordant with 6.25 days reported by Muddaiah et al. [15] Other dominant symptoms were headache, nausea/vomiting and pain in abdomen were similar to those observed by Muddaiah et al. Altered sensorium was observed in 4.21% and 2.7% in their study and ours respectively. [15] The variable manifestations signify the potential of the malaria parasite to affect any organ of the body resulting in organ-specific manifestations.

Hepatomegaly was observed in 39% and 19% cases respectively by Kanodia et al [13] and Kashinkunti et al [16]; our figure of 23.6% lies in between. We observed splenomegaly in 21.8% cases against 53% and 69% reported by Kanodia et al [13] and Kashinkunti et al [16]. The difference can be attributed to the fact that Kanodia et al [13] included patients of severe malaria with acute renal failure; we included all consecutive patients with malaria irrespective of the presence or absence of complications.

Thrombocytopenia was the predominant hematological manifestation of malaria in our study, and was seen in 96.1% cases studied. Similar finding on the high prevalence of thrombocytopenia in patients with P. vivax infection has been described earlier from this region by Kashinkunti et al, [16] Srivastava et al, [17] and De Mast Q [18] et al and observed thrombocytopenia in 63%, 82% and 69% cases of malaria suggesting that thrombocytopenia in malaria, specifically with P. vivax is either a specie- or region- specific finding. The mean platelet counts of $47.0 \pm 59.1 \times 10^3$/cu.mm were comparable to $63 \pm 21.1 \times 10^3$/cu.mm reported in an earlier study. [19] Anemia was observed in 68.1% of all malaria cases concordant with 69% [16] and 86% [20] reported in the past. The high value put forth by Tjitra et al is due to the multi-drug resistant P. vivax infections included in their study that were apparently diagnosed late in the natural history of the disease and hence, were associated with anemia. P. vivax mono-infection was associated in 85.3% cases with anemia while P. falciparum mono-infection was seen in 10.6% in our study. Severe anemia (hemoglobin level < 5gm /dl) was found in 2.7% cases in contrast to 14% and 31.8% respectively reported by Kashikunti et al [16] and Ratan et al [14]. While the former defined severe anemia as hemoglobin < 6 gm/dl causing a higher number of subjects getting included, two-third of the subjects in the latter study were infected with P. falciparum of which 68.7% had anemia. [14] The anemia was predominantly normocytic and apparently hemolysis was the major pathology.

The most common biochemical manifestation of malaria in our study was hyperbilirubinaemia observed in 33.3% cases of which 85.7% were due to P. vivax mono-infection while 7.1% each were due to P. falciparum mono-infection and mixed malarial infections. Hyperbilirubinemia was documented in 34% and 41.8% by Saya et al [22] and Ratan et al [14] in their respective studies. Hyperbilirubinaemia in malaria is multifactorial and may be caused by intravascular hemolysis, disseminated intravascular coagulation (DIC) or hepatocellular dysfunction. Our mean ALT and AST values were higher than normal but lower than a previous study that included only severe vivax infections and hence the disparity [19].

Barcus et al showed the prevalence of malaria associated acute kidney injury as 11.7% in their study on 366 cases, 90.6% being due to P. falciparum infection. [23] Marks et al reported acute kidney injury in 34 to 52 % cases of P. falciparum infections. [24] The mean serum creatinine was 9.2 ± 4.2 mg%, and oligo/anuria was present in 82%; 78% of the patients required hemodialysis. Sixty-four percent of the patients recovered completely, 10% incompletely, and 5% developed chronic renal failure. In our study, acute kidney injury was seen in 24 (21.8%) cases, of which 87.5% and 8.3% were attributed to P. vivax and P. falciparum mono-infections respectively. All cases that underwent dialysis (n=4) were infected by P. vivax.

Shock was observed in 12.4% patients by Ratan et al, of which 88% were due to falciparum mono-infection. [14] While we encountered shock in 13.6% of our cases, 93.3% was attributed to P. vivax mono-infection. Kochar et al reported shock in 7.5 % severe P. vivax infections. [19]

In our study, 97.2% patients survived the illness and were discharged, while 2.7% succumbed. There was no mortality in the dreaded yet small falciparum malaria infected group. Significant association was not seen between Plasmodium species and outcome of the disease (p= 0.394). This is in contrast to other studies where higher mortality was attributed to falciparum than vivax malaria (18.4% vs. 6%). [14] The low mortality in our center is due to the excellent healthcare and intensive care infrastructure availability, use of authentic drugs and adherence to ethics in healthcare. Moreover, we have attained a good experience in the diagnosis and management of complicated cases over the last decade of our existence as the major tertiary referral center of the state catering to a population of over a million from the Garhwal region of Uttarakhand and the densely populated districts of Uttar Pradesh.

The main limitation of our study was the small study group and P. vivax as the dominant organism responsible for most of the cases which gave us no chance to assess the association between complications and comparison between the etiological groups. Cerebral malaria was not observed presumably because of the same reason. Nevertheless, the prospective nature of our study and adherence to WHO definition of severe malaria is the strength of this study.

CONCLUSIONS

This study highlights that acute and complicated malaria is predominantly caused by Plasmodium vivax in north India. A thorough knowledge of complications of malaria can prevent morbidity and mortality in the so-called "benign" albeit "malignant" vivax infection.

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