Original Research Article

Evaluation of Changes in Haematological Parameters in Smear Positive Malaria Cases

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Received: 03-03-2021 / Revised: 22-04-2021 / Accepted: 07-05-2021

Abstract

Background: Malaria causing plasmodia are parasites of blood and hence induce haematological alterations. The haematological changes that have been reported to accompany malaria include anemia, thrombocytopenia and leucocytosis, leukopenia, mild to moderate atypical lymphocytosis, monocytosis, eosinophilia and neutrophilia. **Aim:** To study the changes in haematological parameters in smear positive malaria cases. **Materials and Method:** Total hundred smear positive malaria cases were taken and various hematological parameters and biochemical parameters were studied. **Results:** Out of 100 smear positive cases, P. vivax was positive in 55 cases while P. falciparum was positive in 45 cases. It was seen in 86.67% of falciparum Malaria patients and in 72.72% of vivax Malaria patients. Severe anemia was seen in 9% of patients. Normocytic normochromic blood picture was the most common type in anaemic patients (51.89%). Thrombocytopenia was seen in 71% of the patients. Mild thrombocytopenia was more common and present in 52% of patients while Severe thrombocytopenia was seen in 19% of cases. In falciparum malaria thrombocytopenia was present in 66.66% of the patients while it was present in 74.54% of the patients in vivax malaria. Total Leucocyte Count was normal in 72% of the patients. **Conclusion:** Various haematological findings can help in early diagnosis of malaria which is essential for timely and appropriate treatment which can limit the morbidity and prevent further complications.

Keywords: CBC, Haematological parameters, Malaria, Thrombocytopenia.

Introduction

"Malaria" received its name from Italian as it was believed to arise due to foul air common near marshy areas. More than 100 countries in the world are considered malarious, and more than 2.4 billion of the world's population is at risk. The worldwide annual incidence of malaria is estimated to be about 300-500 million cases. Malaria kills between 1.1 and 2.7 million people annually of which majority are children under five years.[1]

Malaria is a major health problem in India, being one of the biggest burdens in terms of morbidity and mortality among all infectious diseases.[2]

Malaria causing plasmodia are parasites of blood and hence induce hematological alterations. The hematological changes that have been reported to accompany malaria include anemia, thrombocytopenia and leucocytosis, leucopenia, mild to moderate atypical lymphocytosis, monocytosis, eosinophilia and neutrophilia.[3-8] Platelet abnormalities are both qualitative as well as quantitative.

Thrombocytopenia is common occurrence in acute malaria and it is observed in vivaxand falciparum malaria to varying degrees.[6-10] Cases of malaria associated renal and hepatic impairment have been reported from different parts of malaria endemic countries.[11] Hepatic involvement in P. falciparum malaria is not an uncommon presentation and presence of jaundice (bilirubin >3mg/dl) is one of the indicators of severe malaria as defined by the WHO. Jaundice in falciparum malaria may vary from mild to severe and is associated with high incidence of complications and mortality.[12]

There are two major renal syndromes associated with Malaria. (1) A chronic and progressive glomerulopathy that mainly affects African children, classically complicating quartan malaria and (2) ARF associated with falciparum malaria in Southeast Asia, India, and sub-Saharan Africa[13]. Renal impairment is commonly caused by P. falciparum; however, vivax malaria also causes renal impairment. [14]

Hence the present study is undertaken to evaluate the various haematological parameters as well as biochemical parameters affected in malaria and to observe the variations if any, in P. falciparum, P. vivax and mixed infections. The aim of the study is to study the changes in haematological parameters in smear positive malaria cases. To study the changes in biochemical parameters in smear positive malaria cases. To compare these changes in P. vivax and P. falciparum infection

Materials and Method

This prospective study was conducted in the department of pathology at Vardhman Institute of Medical Sciences, Pawapuri. The study was approved by the institutional research and ethical committee. The study was conducted over a period of 6 months time from September 2020 to February 2021.Aninformed and written consent was taken from all the participating subjects prior to the commencement of the study. 100 patients showing smear positivity for one or more species of malaria parasite were included in study. The blood samples of these patients were subjected for following laboratory investigations before starting anti- malarial drugs in all these cases.

Collection of blood

CBC was carried out on Medonic Automated Hematology Cell Counter and following readings were noted.

Hemoglobin (HB%)
HCT
Total leukocyte count (TLC)
Differential leukocyte

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Platelet count.
Biochemical Investigations
Liver function test (LFT)
The patient's samples were processed for Liver function tests including-Serum Bilirubin, AST, ALT with the help of Fully Automated Biochemistry AnalyserSelectra
Kidney function test (KFT)
The patient's samples were processed for Kidney function tests including-Serum creatinine and blood urea with the help of Fully Automated Biochemistry
AnalyserSelectra Peripheral blood smear examination
Peripheral blood smears were prepared using fresh finger prick blood. One drop of blood placed on one side of the slide 1 cm away from end and blood was spread using a spreader slide at angle of 30 degree over the length of slide then slides were left to air dry. Slides were fixed and stained with Leishman stain. Peripheral blood smear examination was done systematically under low, high and oil immersion of microscope for
RBC morphology
Total leukocyte count and differential count
Platelet adequacy
Type of malaria parasite.

Results

Total hundred smear positive malaria cases were taken and various hematological parameters and biochemical parameters were studied. Out of 100 smear positive cases,
P. Vivax was positive in 55 cases while *P. falciparum* was positive in 45 cases. Out of 100 cases, *P. vivax*was the most common observed species. It was seen in 55% of cases. Next common was P. falciparumaccounting for 45% of cases.

Table 1: Malaria cases with different species distribution

Type of parasites	No of patients	Percentage (%)
P. Vivax	55	55
P. Falciparum	45	45
Mixed	0	0
Total	100	100

Most of the cases (57%) were in the adults between 21-40 years age group. There were 20 % of cases below 20 yrs of age group. People of all age groups were seen. Youngest was 1 year old female child with P. vivax infection and oldest was 78 years old female with P. falciparum infection. There were 57 male patients and 43 female patients. Falciparum cases were almost equal in both sexes while Vivax infections were found slightly more in males. Fever was seen in all cases expect one case. Chills and rigor was the next commonest symptom seen in 64% of the cases. Nausea and vomiting was present in 25 cases out of which majority (16 cases) were due to falciparum malaria. Myalgia was present in 14% of cases. Altered sensorium was seen in 3 cases of falciparum and 1 case of vivax infections.

Table 2: Clinical signs in malaria infection

Sign	P. falciparum	P. vivax	Total%
Pallor	29	26	55%
Icterus	7	4	11%
Pedal edema	3	2	5%
Splenomegaly	19	13	32%
Hepatomegaly	11	9	20 %
Hepatosplenomegaly	9	8	17 %
CNS involvement	3	1	4 %

Pallor was the most common clinical sign and was present in 55% of cases. Splenomegaly was present in 32 % of cases. splenomegaly seen in 17% of cases with near equal distribution in Falciparum and Vivax malaria. Icterus was present in 11% of cases. CNS involvement seen in 3 cases of P. falciparum and 1 case of P. vivax.

Investigations

Haemoglobin concentration (Hb%)

Majority of the patients had either mild (40%) or moderate degree (30%) of anemia. Hb Concentration <7 gm% was seen in 9% of the cases; more in Falciparum infection. Haematocrit values less than 20 were seen in 9% of the patients which was slightly more common in Falciparum infection. (11.11%). Most of the patients (68%), showed haematocrit level in the range of 20-35%.

Total leucocyte count (TLC)

Majority of the patients had normal Total WBC count (72%).Reduced WBC count was seen in 18% of the cases and increased counts in 10%, with near equal distribution in vivax and falciparum malaria. Increased WBC count seen in 10% of cases.5 cases of increased neutrophil count were seen, with more in vivax infection (4 cases).Reduced neutrophil count was seen in 12 cases with equal distribution in vivax and falciparum malaria cases. One case of eosinophilia and 6 cases of lymphocytosis were seen in falciparum malaria cases. Two cases of lymphocytosis were seen in vivax infection. Seventy-four cases showed normal differential count.

Table 3: Platelet count

Platelet count	P. falciparum (n=45)	P.vivax(n=55)	Total%
Thrombocytopenia(less than 1.5lakhs/mm^3)	30 (66.66%)	41 (74.54%)	71%
Normal plateletcount (more than 1.5lakhs/mm^3)	15 (33.33%)	14 (25.46%)	29%

Decreased platelet counts were a constant feature of both types of malaria with 71% of cases showing Platelet Count less than 1.5 lakhs/mm^3.Severe Platelet Reduction (<50,000) was seen in 19 cases.
In anemic patients, most commonly RBC's were Normocytic Normochromic (64.55%) followed by Microcytic Hypochromic (29.11%) Microcytic Hypochromic blood picture was seen nearly equal in both falciparum and vivax infection.
Three cases of Macrocytic and two cases of dimorphic blood picture were also seen. Out of the 100 patients, 11 had jaundice. Among them, 7 had falciparum malaria and 4 had vivax malaria. The

diagnosis of malarial hepatitis was made in 3 patients of falciparum malaria on basis of:

Demonstration of Plasmodium infection, at least 3-foldraise in transaminase (especially ALT), with or without conjugated hyper bilirubinaemia, absence of clinical and serological evidence of viral hepatitis and response to antimalarial drugs.

Renal function tests

Serum creatinine level >3.0 mg/dL. It was seen in 1 adult patient. She had Falciparum malaria.

Discussion

Malaria is transmitted by the female anopheles mosquito, causes clinical illness and pathological changes in various body organs with the parasites invading and multiplying in the circulating red blood cells. Malaria causes numerous hematological alterations of which anemia and thrombocytopenia are the most important.

The most common species of malaria in the present study was vivax (55%) followed by falciparum (45%). In studies conducted by Erhart LM et al, Jadhav UM et al, vivax was the most common species while Bashawri LAM et al reported higher falciparum prevalence.[3,16,17] In India, vivax is the most common species encountered followed by falciparum. However, in recent years there has been an upswing in the falciparum cases.Malaria can affect any age group. However, most studies show more of adults as compared to children. The present study had 80 adult patients and 20 patients below age 20 yrs. The mean age of the present study is 30.4 years. Most other studies have mean age groups between 25 and 40. The adult age group is more affected due to their greater mobility and greater risk of exposure due to more outdoor activity. Present study had 57% male patients as compare to 45% female patients. Other studies with comparable results include Jadhav UM et al with 58.3% males, Erhart LM et al with 69% males and Bashawri LAM et al with 75.9% males. [3,16,17]

In present study, Fever was the commonest presenting symptom in 99% of the patients. Chills and rigor was present in 64% of the patients. Nausea and vomiting was seen in 25% of the patients. Headache was seen in 22% of the patients while Altered Sensorium was seen in 4% of patients.

Table 4: Comparison of malaria caused by different species

Type of infection (%)	Bashawri LAM et al[3]	Erhart LM et al[16]	Jadhav UM et al[17]	Present study
P.vivax	39	59	62.17	55
P. falciparum	54.1	38	37.69	45
Mixed	2.33	2	0.04	0

As seen in other studies and our study, Fever is most common symptom. Also chills and rigor, nausea and vomiting, headache are still the common symptoms of malaria. Even though malaria is commonly associated with thrombocytopenia, rash and petechial hemorrhages in the skin or mucous membranes are not the common presentation features.In present study, Pallor was seen in 55% followed by splenomegaly in 32% of cases, hepatomegaly in 20%, Icterus in 11% and CNS involvement in form of seizures and altered sensorium in 4%, and Pedal oedema in 5% of the patients. Variations in different studies may be due to some studies having concentrated only on malarial hepatitis and jaundice in malaria and others on hematological et al had leucopenia in 13.3% of the total malaria cases. Study by Echieverri M et al had 29% cases of leucopenia in vivax malaria cases.[3,23]

While all studies show some changes in the total WBC counts, there is a difference in values. Hence an alteration in the WBC count is not unprecedented either for P. falciparum or P. vivax though the quantum of changes may vary. Present study showed increased neutrophils in 5% of the cases and neutropenia in 12% of the cases.

Similar values are seen in study by Bashawri LAM et al showing 8.3% and 11.6% neutrophilia and neutropenia respectively. Lymphocytosis was seen in the present study in 8% of the cases. Similarly, Biswas et al reported8.5% cases with lymphocytosis.[3,21] parameters only. Anaemia is a frequent finding in malaria cases, particularly in developing nations. In the present study, anaemia (<11.5 gm %) was seen in 79% of the cases. In other studies carried out, Sharma Set al had anaemia in 86.7% of the cases, while in a study conducted by Biswas R et al, 94.4% of the cases had anaemia.[8,21] In study conducted in Saudi Arabia, Bashawri LAM et al had 59.2 % cases showing Anaemia.[3]

Table 5: Comparison of patient age distribution

Studies	Mean age in years
Bashawri LAM et al[3]	25.4
Jadhav UM et al[17]	37.4
Erhart LM et al[16]	28
Present study	30.47

In present study, severe anaemia (<7gm%) was seen in 9% of cases, while study conducted by Bashawri LAM et al had severe anaemia in 5.5% of cases[3]. There is a wide variation in anaemia due to malaria infection depending upon the geographical location of the study. In study conducted by Richard MW et al in London only 15% cases of malaria show anaemia.[22]Studies conducted in developing countries show higher levels of anaemia. In the present study, Leucocytosis was seen in 10% of the cases. Study by Bashawri LAM et al show 7.2% cases with Leucocytosis[3].Sharma SK et al and Biswas R et al show 13.3% and 12.2% cases respectively which are almost similar to the present study[8,21].

More prominent rise (20%) was reported by Ladhani S et al who studied falciparum cases only, and Echieverri M et al studying vivaxcases had only 5% cases of leucocytosis.[15,23] Changes in the WBC are less definite in malaria and there is a wide variation seen among the studies. Usually Total counts in majority of the patients are within normal limits. In the present study 72% of the patients had normal counts. In the present study; increase in Leucocytes in Vivax is seen in 7.8% of the cases while in Falciparum, 10.4% cases show increased Leucocyte count. Leucopenia was seen in 18% of the total cases in the present study. In cases of vivax infection, 19.6% of the cases show fall in Leucocytes while in Falciparum infection 16.6% of the cases show leucopenia. Sharma SK et al observed leucopenia in 6.6% cases in falciparum malaria and Ladhani S et al in 10.2% in falciparum malaria.[8,15]

Table 6: Comparison of clinical signs

Signs	Pallor	Icterus	Splenomegaly	Hepatomegaly	CNS involvement
Farogh A. et al[18]	92%	14%	72%	48%	40%
Muddaiah A et al[19]	11.5%	15.7%	15.7%	4.2%	4.21%
PiplaniS et al[20]	13.1%	6.5%	65.7%	53.9%	-
Present study	55%	11%	32%	20%	4%

In the present study, the percentage of patients showing thrombocytopenia (<1.5 lacs) were 66.66% in case of falciparum malaria and 74.54% in case of vivaxmalaria. The percentage of cases showing thrombocytopenia in falciparum infections and vivax infections varies in different studies. Studies conducted by Bashawri LAM et al and Jhadav UM et al had thrombocytopenia more in Vivax as in the present study while in study conducted by Erhart LM et al, thrombocytopenia is more in cases of falciparum malaria [3,16,17].Thrombocytopenia is a common finding in cases of malaria both vivaxand falciparum as shown by most of the studies conducted. In the present study thrombocytopenia was seen in 71% of all malaria cases. Study conducted by Richards MW et al had thrombocytopenia in 67% of the case.[22]Jaundice was seen in 11% of study group. This Incidence of jaundice is similar to study conducted by Kochar D et al who had 12% of cases with jaundice.24 One had unconjugated hyperbilirubinemia. Majority had conjugated hyperbilirubinemia (10 out of 11).[7] had falciparum malaria and 4 had vivaxmalaria. Three cases fulfilled criteria of Malarial hepatitis. This was almost similar to study of Anand AC et al, who had incidence of malarial hepatitis in 2.4% of cases.[25] Renal failure in the form of acute renal failure was noted in one patient with Falciparum malaria (2.77%). Study by Kochar D et al had the incidence of renal failure to be 2%. No patients with vivax malaria had renal failure.[24]

Conclusion

Malaria is one of the most common infections in Indian Subcontinent. Malaria affects mostly adults with male predominance. Fever, Pallor and Splenomegaly are common clinical features in malaria. Malarial infection causes various haematological and biochemical changes. Anaemia and thrombocytopenia of varying severity are most frequently observed haematological findings however bleeding manifestations are uncommon. In a patient with febrile illness, observation of thrombocytopenia warrants careful search for malaria parasite. P. falciparum is associated with serious complications like Severe anemia, Malarial hepatitis and Renal failure hence P. falciparum infection on suspicion of complication should be further evaluated. Various haematological findings can help in early diagnosis of malaria which is essential for timely and appropriate treatment which can limit the morbidity and prevent further complications.

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Conflict of Interest: Nil Source of support:Nil