International Journal of Clinical and Diagnostic Pathology

ISSN (P): 2617-7226
ISSN (E): 2617-7234
www.patholjournal.com
2024; 7(2): 07-10
Received: 05-01-2024
Accepted: 13-02-2024

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Hematological parameters in cases of malaria: A study at tertiary care hospital in Gujarat

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DOI: https://doi.org/10.33545/pathol.2024.v7.i2a.561

Abstract
Introduction: Malaria is a major public health problem which is associated with high morbidity and mortality. Malaria is caused by protozoan parasite belonging to genus plasmodium.
Aim and Objectives: The aim of the study is to assess hematological parameters in cases of malaria.
Materials and Methods: An observational study was carried out over a period of four months from September 2023 to December 2023 at Narendra Modi Medical College and Sheth L.G. General Hospital, Maninagar, Ahmedabad. Patients with smear positivity for one or more species of malaria were included in study. Cases with history of anti-malarial drug intake and already previously diagnosed malarial cases in the same centre were excluded from the study. A total number of 99 cases were included in present study. All the cases were confirmed to be caused by Plasmodium by demonstration of ring forms (trophozoites), schizonts or gametocytes of the parasite by microscopic examination of peripheral blood smear.
Results: A total of 99 cases were included in present study over a period of 4 months. The age ranged from 7 to 59 years with mean age of 30±11.46 years. A total of 99 cases of malaria were recorded, of which 76% (n=75) had *P. vivax* infection and 24% (n=24) had *P. falciparum* infection. There were no cases of mixed infection. Male predominance (n=56, 56.56%) was noted among total cases of malaria. The most common clinical sign was pallor followed by splenomegaly. Most of the malaria cases had thrombocytopenia. TLC<4000 was noted among 21% (n=21) of malaria cases. 22% (n=22) of malaria cases were anemic.
Conclusion: Malaria mostly affects young adults and has a male preponderance. Hematological alterations encountered in malaria were thrombocytopenia, anemia and leukopenia. A febrile patient presenting with thrombocytopenia and anemia should raise the suspicion of malaria.

Keywords: Malaria, thrombocytopenia, anemia

Introduction

Malaria is a major global health problem in Asia, with an estimated 219 million people affected and causing 4,35,000 deaths globally in 2017. Malaria is caused by a protozoan parasite belonging to genus *Plasmodium*. Out of 172 Plasmodium species known, malarial species known to cause disease in humans include *P. falciparum, P. vivax, P. ovale, P. malariae and P. knowlesi* [1]. The Plasmodium life cycle is very complex and happens in two phases; sexual and asexual, the vector mosquitoes and the vertebrate hosts. The sexual phase of the parasite's life cycle occurs in the mosquitoes. Humans are the intermediate hosts for malaria, where the asexual phase of parasite's life cycle occurs. Female mosquitoes of the genus Anopheles transmit malaria. When an infected female mosquito bites, the parasite in the form of sporozoite enters the human blood and then into enters in to the hepatocytes. The first phase of Plasmodium asexual development happens in the hepatocytes, and then in the erythrocytes. This leads to rupture of erythrocytes by these plasmodium species [2].
Peripheral smear examination and complete hemogram is advised frequently by physicians in patients presenting with fever and chills. Hematological parameters including platelet counts, hemoglobin concentration and total leukocyte count frequently show variation in patients infected by *P. vivax* and *P. falciparum*.
The present study aims to assess abnormal hematological parameters in peripheral smear proven malaria cases.

Materials and Methods

An observational study was carried out over a period of four months from September 2023 to December 2023 at Narendra Modi Medical College and Sheth L.G. General Hospital, Maninagar, Ahmedabad. Patients with smear positivity for one or more species of malaria were included in study. Cases with history of anti-malarial drug intake and already previously diagnosed malarial cases in the same centre were excluded from the study. A total number of 99 cases were included in present study. All the cases were confirmed to be caused by Plasmodium by demonstration of ring forms/trophozoites, schizonts or gametocytes of the parasite by microscopic examination of peripheral blood smear. Further, all these cases were positive for rapid test for malaria PF/PV (PAN) (Biolab Diagnostics (I) Pvt. Ltd.) which utilizes whole blood to detect malarial parasite antigen. This test is sensitive to 5 parasites/microlitre of whole blood and has a specificity of 98%)

Hematological parameters like haemoglobin concentration, total leukocyte count and platelet count were noted from fully automated 5 part cell counter - Mindray. Furthermore, abnormal counts were also manually counted as per standard technique. Repeat platelet counts were done in subjects with severe thrombocytopenia until normal or near-normal values were achieved.

The collected data were recorded in Microsoft excel spreadsheet and were analysed.

Results

A total of 99 cases were included in present study over a period of four months. The age ranged from 7 to 59 years with mean age of 30 years and standard deviation of 11.46 years. A total of 99 cases of malaria were recorded. *P. vivax* infection was noted in 76% (n=75) of patients; while *P. falciparum* infection was noted in 24% (n=24) of patients. There were no cases of mixed infection (Table: 1). Male predominance (n=56, 56.56%) was noted among total cases of malaria. Among *P. vivax* infected patients, 56% (n=42) were males whereas 58% (n=14) of males were seen among *P. falciparum* infected patients. (Table: 2). The most common clinical sign was pallor and was present in 51% (n=50) of cases followed by splenomegaly (n=35, 35%). Hepatomegaly was seen in 34% (n=34) of malaria cases. Hepatosplenomegaly was present in 18% (n=18) of cases. Icterus was present in 11% (n=11) of cases whereas CNS involvement was seen in only 1%(n=1) of cases of malaria(Table: 3). Most of the malaria cases (n=92, 93%) had thrombocytopenia. Normal platelet count was observed in 7% (n=7) of malaria cases. Thrombocytopenia was seen in 93% (n=70) of *P. vivax* infected and 92% (n=22) of *P. falciparum* infected cases (Table: 4). Total leukocyte count (TLC) <4000 was noted among 21% (n=21) of malaria cases. Leukocyte count was normal in 78% (n=79) of malaria cases. Leukopenia was seen in 20% (n=15) and 25% (n=6) of *P. vivax* and *P. falciparum* cases respectively (Table: 5). Anemia was observed in 22% (n=22) of malaria cases, whereas the remaining 78% (n=77) had normal haemoglobin levels. Anemia was observed in 13% (n=10) of *P. vivax* and in 50% (n=12) of *P. falciparum* infected cases (Table: 6).

Table 1: Malaria cases with different species distribution

Type of parasites	No. of patients	Percentage %
P. vivax	75	76%
P. falciparum	24	24%
Mixed	00	00%
Total	99	100%

P. vivax: *Plasmodium Vivax*; *P. falciparum*: *Plasmodium Falciparum*

Table 2: Gender distribution among malaria cases

Type of parasites	Male	Female
P. vivax	42	33
P. falciparum	14	10
Total	56	43

P. vivax: *Plasmodium Vivax*; *P. falciparum*: *Plasmodium Falciparum*

Table 3: Clinical signs in malaria infection

Clinical signs	*P. vivax*	*P. falciparum*	Total
Pallor	28	22	50 (51%)
Icterus	6	5	11 (11%)
Hepatomegaly	15	19	34 (34%)
Splenomegaly	16	19	35 (35%)
Hepatosplenomegaly	8	10	18 (18%)
CNS involvement	00	01	01 (01%)

P. vivax: *Plasmodium Vivax*; *P. falciparum*: *Plasmodium Falciparum*

Table 4: Platelet Count

Platelet Count	*P. vivax*(n=75)	*P. falciparum*(n=24)	Total %
Thrombocytopenia (<1.5 lakhs/mm^3)	70	22	92
Normal Platelet Count (>1.5 lakhs/mm^3)	05	02	07

P. vivax: *Plasmodium Vivax*; *P. falciparum*: *Plasmodium Falciparum*

Table 5: Total Leukocyte Count

Total Leukocyte Count(TLC)	*P. vivax*	*P. falciparum*	Total
<4000	15	06	21
4000 - 10000	60	18	78
Total	75	24	99

P. vivax: *Plasmodium Vivax*; *P. falciparum*: *Plasmodium Falciparum*

Table 6: Hemoglobin concentration

Hb concentration	*P. vivax*	*P. falciparum*	Total
<11 g/dl	10	12	22
>11 g/dl	65	12	77
Total	75	24	99

Hb: haemoglobin; *P. vivax*: *Plasmodium Vivax*; *P. falciparum*: *Plasmodium Falciparum*

Discussion

Malaria presents with varying manifestations. Thus, role of diagnostic modalities is of extreme importance. Malaria causes many alterations in hematological parameters, of which anemia and thrombocytopenia are the most significant. Thrombocytopenia is also seen in patients with acute febrile illness due to viral causes, but its presence is considered as a diagnostic clue for malaria in endemic areas [3].

In present study, the most common species of malaria was *P. vivax* (n=75, 76%) followed by *P. falciparum* (n=25, 24%). This was in concordance with studies conducted by Kumar Manoj *et al*. [3] and Devineni S B, *et al*. [4]. In present study, not a single case of mixed infection was noted which is in concordance with the study conducted by Surve KM *et al*. [5]. In India, the higher prevalence of *P. vivax* malaria is because of variation in climatic conditions, uneven geographic distribution, breeding places of mosquitoes, and genetic resistance to *P. falciparum*.

Male predominance was noted in present study (n=56, 56%). This was in concordance with studies conducted by Kumar Manoj *et al*. [3], Murthy GL, *et al*. [6], Trampuz A, *et al*. [7] and Patel U, *et al*. [8]. This may be because of frequent exposure of males to the risk of acquiring malaria than females as they mostly spend their time outdoors. This increases the risk of exposure to malarial infection.

The mean age of studied patients was 30.27±11.46 years like in study conducted by Kumar Manoj *et al*. [3] having mean age of 33.09±13.53 years. This implies that young adults were most commonly affected which may be explained by the fact of them being active outside of home.

Common clinical signs in decreasing order are pallor (51%), splenomegaly (35%), hepatomegaly (34%), hepatosplenomegaly (18%), icterus (11%) and altered sensorium (1%). A clinical gamut of fever, splenomegaly, and pallor is most often associated with malaria. Clinical signs of pallor and splenomegaly were common in studies conducted by Kumar Manoj *et al*. [3], Farogh A *et al*. [9] and Piplani S *et al*. [10]. Engwerda CR *et al*. [11] observed that splenomegaly in malaria is due to phagocytosis of parasitised red blood cells as an immune response against parasites.

In the present study, thrombocytopenia, anemia and leucopenia were observed in peripheral smear proven malaria patients. Thrombocytopenia was seen in 92 (93%) out of 99 cases. In present study, majority of *P. vivax* cases (n=70, 93%) presented with thrombocytopenia which is similar to Aarti *et al*. [12].

The most constant hematological alteration found in severe malaria is thrombocytopenia. Normal platelet count along with a normal C-reactive protein (CRP) usually excludes the diagnosis. Several mechanisms have been suggested for thrombocytopenia observed in malaria. Evidence of high-molecular-weight Von Willebrand factor in the plasma of patients suggests that thrombocytopenia in malaria is associated with endothelial damage and isolated platelet consumption [13]. Although, exact pathophysiology of reduction in platelet count and increased mean platelet volume is not known, a few hypotheses including coagulation disturbances, splenomegaly, bone marrow alterations, antibody-mediated platelet destruction and oxidative stress have been postulated [14].

Jadhav U M, *et al*. [15]. Found that absence of thrombocytopenia is uncommon in malaria, its presence is not a distinctive feature between the two types of malaria, and severe thrombocytopenia can occur in *P. vivax* and *P. falciparum* infected cases but more commonly in *P. falciparum* infected patients.

Leucopenia was seen in 21% of malaria cases. In malaria, white blood cell counts are generally low to normal [13].

Anemia was seen in 22% of malaria cases. Pathogenesis of malarial anemia is complex and multifactorial. Parasite multiplication leads to decreased hematocrit level due to rupture of RBCs during release of the different stages of maturing parasites. Infected RBCs also undergo oxidative stress, and hemichromes (hemoglobin degradation products) are formed. Hemichromes possess a strong affinity for membrane protein band 3, leading to band 3 oxidation and clustering, similar to normal senescence. Loss of the complement regulatory proteins, including complement receptor type 1 (CR1) and CD55, from the red cell surface, has also been shown to correlate with severe malarial anemia. Another mechanism is defective erythropoiesis. [13].

Severe anemia, defined as a hemoglobin level <7 gm/dl in adults or <5 gm/dl in children, is quite pronounced in *P. falciparum* malaria in contrast to other plasmodia that selectively invade only specific red cell populations. For instance, *P. vivax* and P. ovale tend to invade reticulocytes while *P. malariae* invade more mature forms of erythrocytes [16].

Conclusion

Malaria is a major public health problem in India which mostly affects adults and has a male predominance. Fever, pallor and splenomegaly are common clinical features in malaria. The present study depicts predominant *P. vivax* infected cases over *P. falciparum* cases. The main hematological alterations in malaria infection were thrombocytopenia, leukopenia and anemia. A febrile patient presenting with thrombocytopenia and anemia should raise

the suspicion of malaria. Specific tests can be employed for confirmation.

Acknowledgements
We would like to thank and acknowledge all our senior faculties who have extended their help in completing our study.

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How to Cite This Article
Kartavya JM, Biren JP, Khushi BP, Jinal KP, Swati BP. Hematological parameters in cases of malaria: A study at tertiary care hospital in Gujarat. International Journal of Clinical and Diagnostic Pathology. 2024;7(2):07-10.