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Original Research Article

Hematological changes in patients with malaria in a tertiary care hospital, Multan, Punjab, Pakistan

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ABSTRACT

Background: Malaria is one of the most common human infections and continues to cause significant morbidity and mortality all over the world. To assess and compare the hematological changes in common types of malaria in our patients.
Methods: This observational study included 100 diagnosed malaria patients of Multan Medical and Dental college, Multan both from out patient department (OPD) and in-patient department, between March 2020 and March 2021. The diagnosis of malaria was confirmed by thick and thin film stained with Geimsa's staining for malaria parasite and plasmodium species and the parasite index (MPI) in some cases. Complete blood counts (CBCs) were performed and WBC differential was done on all cases.
Results: The most common type of malarial parasite was *Plasmodium vivax* followed by *Plasmodium falciparum* and 89% of the patient had thrombocytopenia, 70 % anemia, 23% had leukopenia and 10% had raised WBC count. The mean parasite load was 1.275±0.629%, 20 % of the cases showed neutropenia, 40 % had lymphopenia and 40 % showed monocytopenia. Of all the cases 15 % had neutrophilia, 8 % had lymphocytosis and 5 % had eosinophilia. Thrombocytopenia was slightly more in *P. falciparum* (58.69%) than *P. vivax* (30.18%) cases, p>0.05, whereas there was no significant difference in the incidence of anemia in two groups (34.68% vs 33.82%) with p>0.05.
Conclusions: *P. vivax* is the common malarial parasite in our population. Both *P. vivax* and *P. falciparum* can cause marked hematological changes including thrombocytopenia, anemia, lymphopenia and monocytopenia.

Keywords: Malaria, Thrombocytopenia, *Plasmodium vivax*

INTRODUCTION

Malaria has often been considered to fall in the grey zone between parasitology and hematology. A European Textbook of hematology published in the 1930s, defined malaria as a "typical blood disease" with anemia, fever and splenomegaly.[1] Despite global efforts for reducing its transmission, malaria still continues to be a serious and widespread protozoal infection of humans.[2] It is endemic in 107 countries including Pakistan.[3] Globally, around 3.3 billion people are exposed to the risk of malaria, with maximum disease burden in sub-Saharan Africa.[4] More than 40% of the world's population and around 60% of Pakistan's population, lives in malaria endemic regions.[5,6] In Pakistan malaria is mostly found in the provinces of Sindh, Balochistan, Khyber Pakhtunkhwa, and the

federally administered tribal areas.[7] Around 500,000 cases of malaria infection and 50,000 malaria-related deaths are seen each year in Pakistan in spite of an established malaria control programme.[8]

In malaria changes in haematological parameters have been reported earlier. These include anemia, thrombocytopenia and leucopenia or leukocytosis.[9] Splenomegaly, mild-to-moderate atypical lymphocytosis and rarely disseminated intravascular coagulation (DIC) have also been seen.[10] Other hematological reactions to malaria that have been reported are neutropenia, eosinophilia, neutrophilia and monocytosis.[11]

Anemia is one of the most common complications in malaria infection especially in younger children and pregnant women.[12] The pathogenesis of anemia in malaria is multifactorial and includes mechanisms such as destruction of parasitized as well as non-parasitized red blood cells, splenic pooling of red blood cells, oxidative stress induced hemolysis, suppression of marrow hemopoeisis, dyserythropoisis and ineffective erythropoiesis.[13,14] Besides anemia, a reduction in the number of platelets is another one of the more well-known hematologic changes observed in patients with malaria.[15] The pathogenesis of thrombocytopenia is mediated through peripheral destruction, excessive removal of platelets by spleen pooling as well as platelet consumption by the process of disseminated intravascular coagulopathy.[16]

The white blood cell counts and neutrophil counts are usually within normal limits. However on differential count both neutrophilia and neutropenia has been observed in malaria.[17] Alterations in hematological parameters vary with the level of malarial endemicity, background hemoglobinopathy, nutritional status, demographic factors, and also malaria immunity.[18]As malaria is a common parasitic infection in our country, so the purpose of current study was to assess the frequency of hematological complications among malaria cases and its impact on the health of effected patient. We also aimed to see if blood indices can be helpful to support suspicion of malaria in the absence of other more suggestive diagnostic facilities.

METHODS

The study design was prospective observational study. All confirmed cases of malaria presented in Multan medical and dental college/Ibn-e-Sina hospital, Multan between March 2020 and March 2021, were included in the study. A case was defined as a positive thin and/or thick blood film recorded in the hematology department laboratory records. A specially designed form of two sections was used for the purpose of these studies. Basic demographic data: included variables as age, sex, residence. Hematological data: recorded information about complete blood counts (CBC), plasmodium species and the parasite index (MPI) l in some cases. CBC was performed using an automated coulter counter STKS model and WBC differential was done on all the cases. All malaria-positive smears were reviewed by a senior hematologist for confirmation, identification of species, review of smear for platelet count, and in some cases, estimation of parasite level. Normal values (reference ranges) for the hematological findings were based on recommendations by Dacie and Lewis.[13] Data was entered and analyzed using SPSS version 23.0. The numerical variables such as age, hemoglobin, white blood cell count and platelet count were given as mean±SD. The categorical variables such as gender, type of parasite, presence of anemia, presence of thrombocytopenia were given as frequency and percentage. Shapiro-Wilk test was used to check the normality of distribution of data. For comparison of Hb, RBC count, TLC and platelet count among various types of parasitemia ANOVA and Kruskal Wallis test were used. For normally distributed variables such as Hb and RBC count, one-way ANOVA was used. For variables which were not normally distributed such as TLC and Platelet count Kruskal Wallis test was used.

RESULTS

A total of 100 patients fulfilled the inclusion criteria for diagnosis of malaria. The mean age was 25.1±14.8 with minimum of 10 days and maximum 70 years. The majority were males 64% and 36% were females with a male to female ratio of 1.7:1. The most common type of malarial parasite was *Plasmodium vivax* followed by *Plasmodium falciparum*. Mixed infection was 3% and 1% of the cases was due to plasmodium malaria (Figure 1).

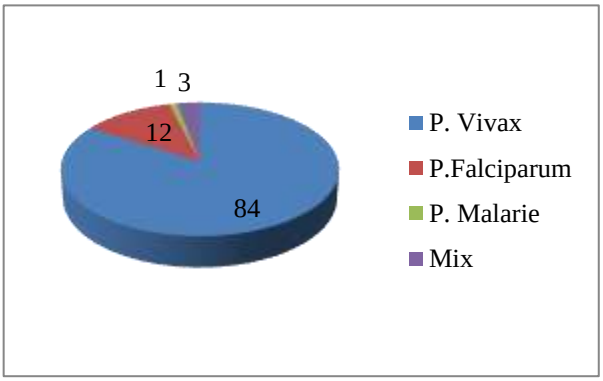

Figure 1: Distribution of malarial parasite type.

Parasite load was also noted among patients. The mean parasite load was 1.3±0.63% with maximum 2.4% and minimum 0.2%. 70 patients were anemic, out of which 58(83%) normocytic, 11(16%) microcytic and only 1(1%) had macrocytic type of anemia. Among these patients 40% had mild, 25% had moderate and 5% had severe anemia. Red blood cell (RBC) count was normal in majority (70%), only 28% patients had low RBC count. The majority of the patients (89%) were thrombocytopenic. The median platelet count was 66.0 (35.3-110.1) $\times 10^9$/ul, with range of 0 to 432×10^9/ul. Of these 37%, 24%, 12% and 11% had moderate, severe,

very severe thrombocytopenia and normal platelet count respectively. The mean platelet volume was within normal limits in 95% patients, high in 4% and low in 1%. The difference of MPV among different parasite infection was found to be statistically insignificant with a p value of 0.986. The white blood cell (WBC) within normal limits in 63% while 23% had leukopenia and 10% had raised WBC count. The median WBC count was 5.2 (3.7-6.9) $\times 10^9$/ul with range of 0.81-28.06 $\times 10^9$ /ul.

Table 1: Hb and RBC comparison among type of parasite.

Parameter		*Plasmodium vivax*	*Plasmodium falciparum*	*Plasmodium malariae*	Mixed infection	P value
Hb	Mean±SD	11.1±2.3	10.5±2.5	15.8	14±2.8	0.033
RBC	Mean±SD	4.1±0.8	4.1±0.9	6.9	5±0.4	0.006

Mean values of different parameters in different malarial type infection were as follow: hemoglobin 11.08±2.34 g/dl in vivax and 10.56±253 in falciparum, WBC count $6.17\pm4.64\times10^9$/ul and $8.61\pm5.27\times10^9$/ul, platelet count $93.96\pm83.18\times10^9$/ul and $44.92\pm41.36\times10^9$/ul, RBC count 4.099±0.88 and 4.08±0.97 x 10^6/ul in *P. vivax* and *P. falciparum* respectively. When different parameters were assessed among different species of malarial parasite infection by using one-way ANOVA, differences in hemoglobin and red cell count were significant with p value of 0.033 and 0.006 respectively. (Table 1) No difference of TLC as well as platelet count was found between parasite types.

Table 2: Parasite type and anemia grade.

Parasite type		Anemia grade				Total
		Mild anemia	Moderate anemia	Severe anemia	No anemia	
Vivax	N	34	22	4	24	84
	%	85.0	88.0	80.0	80.0	84.0
Falciparum	N	5	3	1	3	12
	%	12.5	12.0	20.0	10.0	12.0
Malarie	N	0	0	0	1	1
	%	0	0	0	3.3	1.0
Mix	N	1	0	0	2	3
	%	2.5	0	0	6.7	3.0
Total	N	40	25	5	30	100
	%	100.0	100.0	100.0	100.0	100.0

Pearson Chi-Square value 5.105, P value 0.825

Table 3: Absolute counts of different white blood cells.

Parameter	Mean ($\times10^9$ /ul)	SD	Minimum	Maximum
AbsNeut	5.0	7.33	0.5	56.0
AbsLym	1.4	1.03	0.04	4.87
AbsMono	0.2	0.18	0.04	1.02
AbsEosin	0.1	0.16	0.01	1.00

The severity of anemia when compared among various parasite types showed that among patients with mild anemia 34 (85 %) had *P. vivax* infection, 1 (20 %) had *P. falciparum* and only 1 (2.5 %) had mix infection. Among patients with severe anemia, 4 (80 %) had *P. vivax* infection, and only 1(20%) had *P. falciparum* infection. (Table 2). The absolute white cell counts are given in (Table 3). When the differential white cell counts were analyzed it was found that 20% of malaria patients had neutropenia, 40% had lymphopenia, 40% had monocytepenia and only 2% has eosinopenia. Moreover, neutrophilia was seen in 15% of cases, lymphocytosis in 8% of cases, monocytosis in 1% of the cases and eosinophilia was seen in 5% of the cases.

DISCUSSION

Malaria is one of the most common infections worldwide[1] and is one of the leading causes of febrile illnesses in our part of world. The most frequent manifestation and complication of malaria are changes in hematological parameters. These are changes in red blood cells, platelets and leukocytes and have significant role in malarial pathology.[9]

In our study we found that most common type of malarial parasite was plasmodium *P. vivax* (84%) followed by *P. falciparum* (12%) which was in concordance with the report of National malarial control programme.[7] Mehdi Nateghpouret al in iran reported similar results whereas a

study conducted by Ameekumari Patel in India found more cases of *Plasmodium falciparum* as compared to *Plasmodium vivax*.[19,20] However Zeeba Shamim et al reported results similar to our study with 146 out of 172 patients having *Plasmodium vivax* infection as compared to only 8 patients having *Plasmodium falciparum* infection.[14]

Among the peripheral blood changes anemia is a common presentation as in our case we found that 70% of study subjects had anemia. Anemia was normocytic normochromic in 83% cases while 16% had microcytic hypochromic anemia. Previous studies also showed same results with Latif et al reporting that 97.3% of their malaria patients were anemic and mostly normocytic normochromic anemia has been reported.[20] The pathogenesis of anemia in malaria is multifactorial and is not fully understood. The main mechanisms are destruction of parasitized red blood cells as well as non-parasitized, splenic pooling of red blood cells, oxidative stress induced hemolysis, suppression of marrow hemopoisis, dyserythropoisis and ineffective erythropoiesis.[2,9,10]

The parasitized and non-parasitized red cells are less deformable and are removed in high shear rates of spleen.[21] Increased level of tumor necrosis factor alpha (TNF-α), interleukin 4 (IL-4) and interferon gamma (IFN-γ) are proposed to cause bone marrow suppression and ineffective erythropoiesis. Anemia of chronic disorder due to failure of marrow to utilize the iron due to raised level of IL-6 and TNF-α. Another proposed mechanism is imbalance in red cell surface marker such as chemokine receptor-1.[22]All these mechanism lead to the development of anemia.[23] In our study most common hematological abnormality was thrombocytopenia (89%). Previous studies had shown that thrombocytopenia is a characteristic finding of malarial infection and is more common than anemia.[15] We observed that thrombocytopenia is more frequent finding in *P. vivax* infection. Saravu et al reported that the prevalence of thrombocytopenia was similar amongst both infection of vivax and falciparum malaria, but patients with severe falciparum malaria had a significantly lower platelet count compared to the non-severe falciparum malarial patients.[24] Other studies have reported lower platelet counts among patients infected with *P. falciparum* in comparison to those of *P. vivax*.[25]

The pathogenesis of thrombocytopenia is multifactorial. Demonstration of *P. vivax* within platelets and a direct lytic effect of the parasite on the platelets has been suggested by some.[26] Another mechanism may be the generation of immune complexes by malaria antigen which might lead to sequestration of the injured platelets by macrophages in the spleen.[27] Platelet consumption by the process of disseminated intravascular coagulation (DIC) as well as antibody mediated platelet destruction is another mechanism causing thrombocytopenia.[28] WBC count was within normal limits in majority of cases (63%) in our study and same finding was reported in many previous studies.[29] Differential leukocyte count showed neutropenia in 20% cases, neutrophilia in 15%, lymphopenia in 40% and lymphocytosis in 8% cases. Differential leukocyte count is also usually reported to be within normal limits, however both neutrophillia and neutropenia has been observed in malaria.[15,30] In this study 40% cases showed monocytopenia on differential leucocyte count. This is in agreement with a study published by Prasad et al who found neutrophilia and monocytopenia in malaria patients.[31]

CONCLUSION

Present study concludes that significant hematological changes occur in malaria infection constituting mainly anemia and thrombocytopenia. Changes in the white blood cell are less pronounced with the total count being normal mostly but differential count showing lymphocytopenia and monocytopenia in 40% cases. It would be worthwhile to carry out prospective studies on other hematological variables such as coagulation parameters, bone marrow changes and the direct coomb's test to further improve our understanding of the anemia in malaria.

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REFERENCES

1. Bashawri LA, Mandil AA, Bahnassy AA, Ahmed MA. Malaria: hematological aspects. Ann Saudi Med. 2002;22:372-6.
2. Rodrigues-da-Silva RN, Lima-Junior Jda C, Fonseca Bde P, Antas PR, Baldez A, Storer FL, et al. Alterations in cytokines and haematological parameters during the acute and convalescent phases of Plasmodium falciparum and Plasmodium vivax infections. Mem Inst Oswaldo Cruz. 2014;109(2):154-62.
3. Latif I, Jamal A. Hematological changes in complete blood picture in paedriatric patients of malaria caused by plasmodium vivax and falciparum. J Ayub Med College Abbottabad. 2015;27(2):351-5.
4. Khan W, Zakai H. Clinico-pathological studies of Plasmodium falciparum and Plasmodium vivax-malaria in India and Saudi Arabia. Acta Parasitol. 2014;59:206-12.
5. Akhtar S, Gumashta R, Mahore S, Maimoon S. Hematological changes in malaria: a comparative study. IOSR-JPBS. 2012;2:15-9.

6. Khattak AA, Venkatesan M, Nadeem MF. Prevalence and distribution of human Plasmodium infection in Pakistan. 2013;12:297.
7. Kakar Q, Khan MA, Bile KM. Malaria control in Pakistan: new tools at hand but challenging epidemiological realities. 2010;16:54-60.
8. Mukhtar M. Killer number one: the fight against malaria: malaria strategy lags behind the global goals, Humanitarian news and analysis a service of the UN Office for the Coordination of Humanitarian Affairs. Nairobi. 2006.
9. Arévalo-Herrera M, Lopez-Perez M, Medina L. Clinical profile of Plasmodium falciparum and Plasmodium vivax infections in low and unstable malaria transmission settings of Colombia. Malaria J. 2015;14:154.
10. Al-Salahy M, Shnawa B, Abed G, et al. Parasitaemia and its relation to hematological parameters and liver function among patients malaria in Abs, Hajjah, Northwest Yemen. Interdis Perspect Infect Dis. 2016.
11. Muwonge H, Kikomeko S, Sembajjwe LF, Seguya A. Namugwanya C. How reliable are hematological parameters in predicting uncomplicated plasmodium falciparum malaria in an Endemic Region?. ISRN Tropical. 2013:2.
12. Menendez C, Fleming AF, Alonso PL. Malaria-related anaemia. Parasitol. 2000. 16:469-76.
13. Kai OK, Roberts DJ. The pathophysiology of malarial anaemia: where have all the red cells gone?. BMC Med. 2008;6:24.
14. Jairajpuri ZS, Rana S, Hassan MJ, Nabi F, Jetley S. An Analysis of Hematological Parameters as a Diagnostic test for Malaria in Patients with Acute Febrile Illness: An Institutional Experience. Oman Med J. 2014;29(1):12-7.
15. Kotepui M, Piwkham D, Phun Phuech B. Effects of malaria parasite density on blood cell parameters. PLoS One. 2015;10:0121057.
16. Kotepui M, Phunphuech B, Phiwklam N, Chupeerach C, Duangmano S. Effect of malarial infection on haematological parameters in population near Thailand-Myanmar border. Malar J. 2014;13:218.
17. Khan AS, Malik SA. Haematological changes in falciparum malaria and tumor necrosis factor. J Pak Med Assoc. 1996;46:198-200.
18. Erhart LM, Yingyuen K, Chuanak N, Buathong N, Laoboonchai A, Miller RS, Meshnick SR, Gasser RA Jr, Wongsrichanalai C. Hematologic and clinical indices of malaria in a semi-immune population of western Thailand. Am J Trop Med Hyg. 2004;70(1):8-14.
19. Nateghpour M, Hosseininasab A, Farrokhnia M, Dastouri F, Alidoosti K, Sadequi D, Ahmadi A. Species-dependent Clinical Findings of Malaria Caused by Various Plasmodia in an Endemic Area of Kerman Province, Southeastern Iran. Iran J Public Health. 2017;46(4):525-529.
20. Patel A, Awoke N, Arota A. Profiles of hematological parameters in Plasmodium falciparum and Plasmodium vivax malaria patients attending Tercha General Hospital, Dawuro Zone, South Ethiopia. Infect Drug Resist. 2019;12:521-7.
21. Roberts DJ, Casals-Pascual C, Weatherall DJ. The clinical and pathophysiological features of malarial anaemia. Curr Top Microbiol Immunol. 2005;295:137-67.
22. Chang KH, Stevenson MM. Malarial anaemia: mechanisms and implications of insufficient erythropoiesis during blood-stage malaria. Int J Parasitol. 2004;34(13-14):1501-16.
23. Ghosh K, Ghosh K. Pathogenesis of anemia in malaria: a concise review. Parasitol Res. 2007;101:1463-9.
24. Saravu K, Docherla M, Vasudev A, Shastry BA. Thrombocytopenia in vivax and falciparum malaria: an observational study of 131 patients in Karnataka, India. Ann Trop Med Parasitol. 2011;105(8):593-8.
25. Pain A, Ferguson DJ, Kai O, Urban BC, Lowe B, Marsh K, Roberts DJ. Platelet-mediated clumping of Plasmodium falciparum-infected erythrocytes is a common adhesive phenotype and is associated with severe malaria. Proc Natl Acad Sci. 2001;98(4):1805-10.
26. Kumar J, Arshad AR. Thrombocytopenia in malaria: can platelet counts differentiate malaria from other infections? J Coll Physicians Surg Pak. 2015;25(1):31-4.
27. Coelho HC, Lopes SC, Pimentel JP. Thrombocytopenia in Plasmodium vivax malaria is related to platelets phagocytosis. PLoS One. 2013;8(5):e63410.
28. Maina RN, Maina RN, Walsh D, Gaddy C, Hongo G, Waitumbi J, Otieno L, Jones D, Ogutu BR. Impact of Plasmodium falciparum infection on haematological parameters in children living in Western Kenya. Malar J. 2010;9(Suppl 3):S4.
29. McKenzie FE, Prudhomme WA, Magill AJ, Forney JR, Permpanich B, Lucas C, Gasser RA Jr, Wongsrichanalai C. White blood cell counts and malaria. J Infect Dis. 2005;192(2):323-30.
30. Dale DC, Wolff SM. Studies of the neutropenia of acute malaria. Blood. 1973;41(2):197-206.
31. Prasad PL, Rai PL, Hussain MS, Karoli R, Shakya S, Gupta N, et al. Clinical profile of malaria at a tertiary care teaching hospital in North India. Trop Parasitol. 2021;11(1):25-30.

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