ORIGINAL ARTICLE

HEMATOLOGICAL PROFILE OF PLASMODIUM VIVAX AND PLASMODIUM FALCIPARUM INFECTED PATIENTS COMPARED WITH CONTROL GROUP IN HAYATABAD MEDICAL COMPLEX, PESHAWAR

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ABSTRACT

Objective: To compare hematological parameters of P. falciparum and P. vivax infected patients with the control group.

Material & Methods: It was cross-sectional study conduct at Hayatabad Medical Complex, Peshawar. One hundred and seventy-two laboratory confirmed cases of malaria were included in the study. Hematological profile of subjects infected with P. vivax, P. falciparum, and control group were compared.

Results: The platelets, red blood cells, and leucocytes count, hemoglobin level and mean corpuscular hemoglobin concentration (MCHC) were considerably decreased in both P. falciparum and P. vivax infected subjects in contrast to control group, whereas reduction in lymphocyte count was observed only in P. vivax infected patients. Monocytes count and hematocrit (HCT) was considerably higher in P. vivax infected subjects. However, no significant difference was noticed in the neutrophil and eosinophil count and level of mean corpuscular hemoglobin (MCH) and mean corpuscular volume (MCV) among the three groups.

Conclusion: Patients infected with malaria showed essential variations in some hematological parameters with low platelet count and hemoglobin concentration being the two most significant indicators of malaria infection.

Key Words: P. vivax, P. falciparum, Blood, Malaria.

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INTRODUCTION

Malaria is common in Pakistan, India, and Bangladesh.[1] The prevalent species of malaria in Pakistan are P. vivax and P. Falciparum.[2] About 60% of Pakistanis population is living in those regions where the malarial infection is endemic.[3] Malaria is a protozoal disease primarily caused by parasitic infection of the genus plasmodium. Plasmodium is an obligate intracellular parasite that is capable to infect and multiply within red blood cells (RBCs).[4] This disease is transmitted to humans by the bite of certain species of infected female Anopheles mosquito. It can also be transmitted by blood transfusions, which is uncommon.[5] Infected needles and from mother to fetus during pregnancy are also the other routes of transmission.[6] There are five species of plasmodium, which cause malarial infection in humans; Plasmodium vivax (P. vivax), Plasmodium falciparum (P. falciparum), Plasmodium malariae (P. malariae), Plasmodium ovale (P. ovale) and Plasmodium knowlesi (P. knowlesi).[7]

P. falciparum is the most common pathogenic and deadly parasite causing malaria in humans among all species.[4] It is responsible for about 90% deaths of the total malaria cases.[8]

Common complications of malaria are hematological changes.[9] The degree of reduction in blood counts varies with the type of malarial infection. The alterations in hematological parameters due to malarial infection include anemia, thrombocytopenia, atypical lymphocytosis and less commonly disseminated intravascular coagulation.[10] Early and common sign of malarial infection is thrombocytopenia. [11] Though anemia in the malarial infection is hemolytic in nature, hematopoietic response is blunted, as evidenced from disproportionate reticulocytes counts, reduced white blood cells (WBCs) and platelets counts indicating some problem with the process of hematopoiesis in the bone marrow Leucopenia, leukocytosis, neutropenia, neutrophilia, eosinophilia, and monocytosis also have been reported. These changes depend on the endemicity of malaria, nutritional status, hemoglobinopathy backgrounds, demographic factors and also individual immunity against malaria. [10,12,13] It is well known that infection of malaria can cause structural, morphological and biochemical changes in the RBC membrane to enable growth, compartmentalization, cell differentiation and increased uptake of nutrients and survival of parasites. [14]

Changes in the parameters of complete blood count induced by malaria are reported from around the globe including Kuwait, Saudi Arabia and Pakistan. [15] It includes thrombocytopenia, anemia, neutrophilia, neutropenia, immature neutrophils, toxic granulation of neutrophil, lymphocytosis, lymphopenia, monocytosis, eosinophilia, and post-treatment eosinophilia.[16] The objective was to compare the hematological parameters between P. falciparum and P. vivax infected subjects with the control group.

MATERIAL AND METHODS

It was a cross-sectional study carried out over six months in Hayatabad Medical Complex Peshawar. Permission was taken from the head of the pathology department. The sample size was calculated considering 18% prevalence of malaria, 5% marginal error with 95% confidence interval.[17] In this study, 172 positive malarial patients were select randomly. Two-milliliter blood was taken from the patients in the EDTA tube using an aseptic technique. Both thick and thin slides were prepared from blood and stained with Giemsa stain. These slides were observed using the oil emersion lens. Informed consent was taken from the patients before including them in the study. SPSS-22 (SPSS Inc. 2007) was used for statistical analysis. Descriptive statistics were used to check data for entry errors. The data was checked for normalization using frequency histograms. All tests were performed at the significance level of 0.05. One-way analysis of variance (ANOVA) with Tukey's Honest Significance Difference (HSD) multiple comparisons were used to test the differences in continuous measures among more than two categories.

RESULTS

A total of 172 malaria patients and 50 healthy individuals were selected as the control group, were included in the study. Among 172 patients, 107 (62.2%) were males and 65 (37.8%) were females. P. vivax was predominant species present in 161 (93.60%) out of which 99 (61.49%) were males and 62 (38.51%) were females, while P. falciparum was found in 11 patients, 08 (72.72%) were males and 03 (27.28%) were females.

Sixty-six percent (66.4%) of the patients affected with P. vivax had a normal level of hemoglobin, whereas 33.6 % of patients were anemic. Similarly, in P. falciparum affected patient 63% of patients had a normal level of hemoglobin (HB) and 37% of patients were anemic (Table 1). Most of the malaria patients had low platelets count both in P. vivax and P. falciparum affected patients (Table 2). In P. vivax affected patients, the count of monocytes was significantly raised as compared to the control group p=0.000. 109 (67.70%) patients had normal monocytes count, 52 (32.29%) had monocytosis. However, in the case of eosinophil, no significance was found among the three groups. (Table 3).

RBC count was considerably decreased in P. falciparum infection in contrast to the control group both in males and females (p=0.011). Similarly, RBC count was reduced in P. vivax

affected subjects as compared to the control group p=0.000, however, these were within the normal reference range in all the three groups. The mean RBC count in P. vivax, P. falciparum, and control group are 4.52±0.65/µl, 4.06±0.79/µl, and 4.93±0.48/µl respectively.

There was no significant difference in mean corpuscular hemoglobin (MCH) among the groups, but mean corpuscular hemoglobin concentration (MCHC) was considerably decrease in P. vivax and P. falciparum as compared to control group (Table 4).

Table 1: Mean HB (g/dL) of P. vivax, P. falciparum infected and control group

Age (Years)	P. vivax Hemoglobin Level		P. falciparum Hemoglobin Level		Control Hemoglobin Level	
	Male	Female	Male	Female	Male	Female
1-10	9.9±1.8	10.6±1.2	11.3±0.3	10.9±0.3	12.8±0.9	14.2±1.2
11-20	11.6±1.6	11.2±1.4	11.9±2.0	11.3±2.0	14.2±1.0	13.1±0.9
21-30	12.7±1.4	11.7±1.6	11.1±3.9	10.4±2.0	14.95±0.9	12.4±0.3
31-40	12.4±2.2	11.2±1.9	-----	12.1±0.9	14.8±0.3	13.0±0.7

Table 2: Comparison of platelets count among P. vivax, P. falciparum and control group

Age	P. vivax		P. falciparum		Control	
	Male	Female	Male	Female	Male	Female
1-10	122000±51692.67/µl	99300±45000/µl	146400±79000/µl	148000±7070/µl	319000±51300/µl	248000±1732/µl
11-20	105800±37426.65/µl	144000±76400/µl	118000±66240/µl	128000±1415/µl	261800±79800/µl	309000±34390/µl
21-30	113000±54000/µl	139000±81790/µl	110300±77800/µl	148500±38890/µl	229400±65100/µl	197200±55700/µl
31-30	106600±47000/µl	104600±48160/µl	00	94400±55950/µl	191100±30315/µl	252600±65400/µl

Table 3: Differential Leucocytes Count of P. vivax and P. falciparum

Parameters	P. vivax	P. falciparum	Control group	P value*	P value**
Neutrophil	58.34±8.47%	58.81±12.84%	62.03±15.27%	0.17	0.989
Lymphocytes	24.47±13.10%	30.0±12.77%	31.62±7.63%	0.000	0.917
Monocyte	9.42±4.41%	8.09±2.06%	7.14±2.29%	0.000	0.485
Eosinophil	3.09±1.76%	2.81±1.40%	2.9±1.40%	0.651	0.993

Table 4: RBC indices of P. vivax and P. falciparum infected patients and control group

Parameters	P. vivax	P. falciparum	Control group	P value*	P value**
MCH	27.10±5.67pg	27.09±2.56pg	28.31±1.61pg	.058	.373
MCHC	31.52±2.82g/dL	30.99±1.68 g/dL	32.97±1.61 g/dL	.000**	.009*
MCV	84.92±6.11fL	86.34±5.63Fl	86.51±4.41fL	.116	.993
HCT	42.64±4.16%	37.15±6.68%	38.63±5.83%	0.000**	0.062

P-value*. P-value when P. vivax and control groups were compared, P value**. P-value when P. falciparum and control groups were compared.

DISCUSSION

In this study, several significant changes were observed in hemoglobin, platelets and differential leucocytes count. Hemoglobin was considerably reduced in the P. falciparum group and P. vivax group in contrast to the control group. This result is parallel with the study done by Maina RN et al., on Plasmodium infection.[9] A similar study was conducted by Igbeneghu and Odaibo in Southwestern Nigeria and showed similar results with the current study.[18] A significant reduction was found in the number of RBCs in P. vivax and P. falciparum-infected patients as compared to the control group but these values were within the normal range ($p < 0.05$). These results are similar to the study done by Igbeneghu and Odaibo in 2013 and the study done by Ashis.[18,19]

Thrombocytopenia was more severe in P. Vivax than the P. falciparum group. Many studies like Lee et al., and Oh et al., reported that there has been an association of thrombocytopenia and malaria which shows comparable results with the recent study.[20-23] Mean WBC count was considerably lower in the P. falciparum group in contrsst to P. vivax group but the average WBC count was normal in both P. vivax and P. falciparum affected patients. Our study is consistent with a study that has reported that the total WBC count is within normal range.[24,25] Except in a few studies, where there was evidence of leucopenia.[26] In another study done in India in the malaria-endemic area, where there was also evidence of leucopenia.[27] This difference might be due to the inclusion and exclusion criteria. In the recent study, all those patients who were suffering from chronic malaria, anemia, and nutritional insufficiency and having a history of leukemia were excluded from the study. Patients with typhoid fever and dengue fever were also excluded. For example, one of the studies done by Abdool Gaffar et al., reported that typhoid fever also causes leukopenia.[28]

Generally, in acute malaria, the lymphocyte count fluctuations were seen in various studies. Lymphopenia may develop in non-immune adults and kids in endemic zones.[25,29] However, the results suggested that the differential lymphocyte count was normal in both P. vivax and P. falciparum group. Some studies suggest that these components for transient lymphopenia are due to the free streaming circulation system to the endothelial coating of the vessels to adhere, distribution of lymphocytes in tissues and besides destruction of lymphocytes due to Fas-induced apoptosis.[30–32] A similar study was performed in Thailand-Myanmar which showed the same results about lymphocytes.[33] Monocytosis was the steady hematological finding in various investigations of acute malaria. [9,10,34] However, in the present study, the differential monocytes count was within normal ranges. The result of monocytes count is similar to a study done by M Kotepui.[33] Neutrophils count in P. vivax and P. falciparum group was within reference range but significantly reduced in P. vivax group as compared to the control group, whereas no significant difference was observed between P. falciparum and control group. These results are consistent with the studies done by Akhtar S et al., in Nagpur, Maharashtra and in Kolkata, India by Ashis Kumar et al., [10,19] However, few studies exhibited neutropenia which might be the due to expanded margination and sequestration of neutrophils because of the increased expression of adhesion molecules of cell [ICAM—1 and VCAM—1] happened in acute malaria.[31]

The result of eosinophils count is also comparable with other studies.[19,35] The value of MCHC was considerably decreased in P. vivax group and P. falciparum group compared to the control group, which is consistent with the study done in Tamil Nadu, India.[19] There was no considerable difference in MCH value between P. vivax group and the control group. Similarly, no significant difference was observed between the P. falciparum group and the control group. MCH is within reference level but low in both P. vivax and P. falciparum groups as compared to the control group. MCV was within the reference range and no significant difference was found among them. The results of MCV and MCH are comparable with the Myanmar study.[33]

CONCLUSION

We observed that hematological changes, for example, thrombocytopenia, anemia, and varying leucocyte count demonstrated a statistically significant connection with malaria infection. We presumed that routinely utilized lab findings, for example, hemoglobin, leukocyte, and platelet counts can give an analytic piece of information in a patient with acute febrile illness in endemic territories, consequently increasing the likelihood of effectively diagnosing malaria and improving treatment initiation promptly. It is intriguing to additionally assess a large number of specimens estimate and recognize, and also compare the difference between malaria-affected and non-infected cases.

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