Study of Hematological Parameters In Plasmodium Vivax Malaria In Mardan, Khyber Pakhtunkhwa.

Salva Shah¹, Mohtasim Billah¹, Nazish Shinwari², Adeel Basharat³, Maheen Shah⁴

ABSTRACT

Background: Malaria is among the most prevalent infections of the world. It is caused by plasmodium species which are parasites of blood thus inducing hematological alterations.

Objective: This study was done to observe the complete hematological profile in Plasmodium vivax induced malaria patients and the associated microscopic findings in these patients.

Materials & Methods: This was a cross-sectional study done in Mardan Medical Complex, Mardan during July to November, 2018. 100 cases of Plasmodium vivax malaria whose diagnosis were confirmed through Giemsa stained thick and thin blood films were taken through consecutive sampling. Hemoglobin (Hb), hematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), red cell count (RCC), total leucocyte count (TLC) and platelet count were done using hematology analyzer and detailed microscopy was done to find other morphological changes in red cells.

Results: Age range of patients was 4 to 66 years. 60% of positive cases were males and 40 % females. Thrombocytopenia was the most common hematological abnormality present in 70% patients followed by anemia which was seen in 58% of patients. Microcytic hypochromic was the commonest blood picture in anemic patients. Leucopenia was present in 3% of patients only while leukocytosis in 15% of patients.

Conclusion: Thrombocytopenia and anemia were the most common associated abnormalities in P. vivax malaria whereas microcytosis and hypochromia were the most common microscopic findings.

Keywords: Malaria, Plasmodium vivax, thrombocytopenia, anemia

INTRODUCTION

Malaria is among the most prevalent infectious diseases of the world. The existence of this disease can be traced back to 2700 years BC in China and historians ¹. The causative agent of the disease is a parasitic unicellular organism of genus Plasmodium which gets entry into the human blood stream through the bite of female anopheles mosquito. Traditionally four species of plasmodium, including Plasmodium falciparum, P. vivax, P. ovale and P. malariae have been known to be infectious to humans ². According to the World Health Organization (WHO) estimates, 40% of the world’s population is at risk of developing malaria. Global incidence of 300 - 500 million cases per year has been reported by studies with an associated two million deaths per annum ³. Likewise, in Pakistan, the disease plays havoc with lives of millions per year and local literature reveals higher mortality rates among infants, children, and pregnant women ⁴,⁵. The predisposing factors include unrestrained migration of Afghan refugees, urbanization, an expansive irrigation network, uncontrolled growth rate of population, sudden environmental changes due to excessive and monsoon rains, earth quakes, floods, and water development projects that made the environment of Pakistan favorable for persistence of malarial species.¹ The Plasmodium vivax malaria is prevalent in many regions of the world accounting for more than half of the cases in Asia and Latin America ⁷. The research done on malaria in the past was confined to P. falciparum infection as compared to P. vivax due to its high mortality rate in different regions of the world. Nowadays, P. vivax has been accepted as an emerging threat to human, as it is considered a major cause of malaria morbidity outside Africa, especially Asia and the Americas. In addition, P. vivax is an alarming threat to almost 40% of the world’s population, including 132 to 391 million clinical infections annually ⁸,⁹.

In view of the paucity of data from Pakistan, we have attempted to throw some light on the laboratory profile of patients suffering from malaria caused by P. vivax by analyzing and correlating various hematological indices and comparing them with the available literature worldwide.

References:
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2. Khyber Medical College, Peshawar
3. Lady Reading Hospital, Peshawar
4. Abottabad International Medical College, Abbottabad

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265
MATERIAL & METHODS
This was a cross-sectional observational study conducted in Pathology Department Mardan Medical Complex, Mardan during July to November, 2018. Total 100 patients suffering from Plasmodium vivax malaria were selected through consecutive sampling technique. The inclusion criteria was all those patients whose diagnosis of malaria was confirmed by thin and thick blood films stained with Giemsa stain for malarial parasite. Exclusion criteria were those malarial patients who were known to have chronic illnesses as tuberculosis and malignancy. All malaria positive cases were reviewed by hematologist for confirmation, identification of species and review of smear for platelets count and other hematological changes. Blood samples of all the patients referred for peripheral blood smear were drawn through venipuncture by professional staff into a 3 mL tube with ethylenediaminetetraacetic acid (EDTA) and were also analyzed for malarial parasites with conventional microscopy. Hemoglobin (Hb), hematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), red blood cell (RBC) count, total leukocyte count (TLC), and platelet count were determined by using a hematology analyzer CELL-DYN Ruby.

Various hematological indices were analyzed using the WHO criteria. For hemoglobin, the cut-off criterion indicating anemia was 12 g/dL for females and 13 g/dL for males. Hematocrit was considered to be abnormal at values < 36% for females and < 41% for males. Likewise, RBC count for males was considered normal in the range of 4.2 - 5.8 $\times 10^6$/cubic millimeter (mm$^3$) and between 3.6 - 5.6 $\times 10^6$/mm$^3$ was considered within normal limits for females. The cut-off values for RBC indices indicating anemia were as follows: MCV < 80 femtoliters (fL), MCH < 27 picograms (pg), and MCHC < 32 grams per deciliter (g/dL). Similarly, platelets < 150 $\times 10^3$/cmm and TLC values < 4 $\times 10^3$/mm$^3$ were considered abnormal.

The data of positive cases of malaria and laboratory findings were recorded on predesigned proformas designed for this purpose.

All the study variables like age, sex, hemoglobin level, total leucocyte count and platelet count were analysed for descriptive statistics like percentages, mean and standard deviation was calculated by using SPSS version 20.

### RESULTS
A total of 100 smear-positive P. vivax patients were included in this study in which 60% were males and 40% were females. The mean age of patients was 28.4 ± 19.4 years. Age range in this study was 4 to 66 years. Most patients were in the age 18 to 50 years.

The minimum Hb noted was 4.9 gm/dl and maximum was 19.4 gm/dl with mean of 11.5 ± 2.46 gm/dl. The normal hemoglobin levels were present in 42% cases whereas 58% were anemic. Hemoglobin levels of less than 8 gm/dl is labelled as severe anemia and in this study only 7% of patients of malaria had severe anemia.

Red cell count ranged between 1.87 to 6.21 $10^6$/µL. Mean red cell count was 4.2 $10^6$/µL. Hematocrit (HCT) levels were in the range of 15-56.2 % and mean was 35.36 %.

TLC was minimum 1.97 and maximum 19.70 $10^3$/cmm with the mean value of 7.98 $10^3$/cmm. Majority of patients (82%) had leukocyte counts within reference limits. Leucopenia was found in 3% whereas leukocytosis was present in 15% patients.
The normal platelet count is 150,000 to 450,000/cmm. The lowest platelet count noted was 22000/µl whereas the highest was 540000/µl. Mean platelet count observed was 136000/µl. Total 30% patients had normal platelet counts, 70% had thrombocytopenia and none had reactive thrombocytosis. 59% of patients had mild thrombocytopenia (50-150000/µl), 11% had moderate thrombocytopenia (20-50000/µl) while severe thrombocytopenia (less than 20,000) was not present in any of these patients.

Bicytopenia was found in 53% whereas pancytopenia in 3% of cases.

Haemoglobin and total leucocyte count in malaria in all age groups.

**DISCUSSION**

The age range of patients infected with Plasmodium vivax in our study was 4 to 66 years. The highest number of patients (71%) were in the age group of 18-50 years. This observation was in accordance with the study done by Aundhakhar *et al* in 2015 who showed that 78% of malaria patients were in the age range of 18 to 50 years. Studies done by Suryawanshi *et al* and Muddhiah *et al* also showed that maximum number of patients reported were in young and middle age group. The reason for this might be that people of this age group are more active outdoors.

60% of positive cases were male and 40% female with the male to female ratio 1.5:1. Our study showed male preponderance which was consistent with studies done previously by Wasnik *et al* and Deshwal.

### Table 1: Frequency of Full Blood Count Parameters

<table>
<thead>
<tr>
<th>PARAMETER</th>
<th>RANGE</th>
<th>PERCENTAGE OF PATIENTS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hb (g/dl)</td>
<td>Less than 4</td>
<td>0%</td>
</tr>
<tr>
<td></td>
<td>4.1-8</td>
<td>7%</td>
</tr>
<tr>
<td></td>
<td>8.1-12</td>
<td>51%</td>
</tr>
<tr>
<td></td>
<td>Greater than 12</td>
<td>42%</td>
</tr>
<tr>
<td>TLC (×10³/µl)</td>
<td>Less than 2</td>
<td>1%</td>
</tr>
<tr>
<td></td>
<td>2.1-4.0</td>
<td>2%</td>
</tr>
<tr>
<td></td>
<td>4.1-8.0</td>
<td>55%</td>
</tr>
<tr>
<td></td>
<td>8.1-11.0</td>
<td>27%</td>
</tr>
<tr>
<td></td>
<td>11.1-15.0</td>
<td>13%</td>
</tr>
<tr>
<td></td>
<td>15.1-20.0</td>
<td>2%</td>
</tr>
<tr>
<td>Platelet count</td>
<td>Less than 20,000/µl</td>
<td>0%</td>
</tr>
<tr>
<td></td>
<td>20,000-50,000/µl</td>
<td>11%</td>
</tr>
<tr>
<td></td>
<td>50,000-150,000/µl</td>
<td>59%</td>
</tr>
<tr>
<td></td>
<td>Greater than 150,000/µl</td>
<td>30%</td>
</tr>
</tbody>
</table>

### Table 2: Frequency of Microscopic Findings on Peripheral Smear

<table>
<thead>
<tr>
<th>MICROSCOPIC FINDING</th>
<th>FREQUENCY</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hypochromia</td>
<td>61%</td>
</tr>
<tr>
<td>Microcytosis</td>
<td>29%</td>
</tr>
<tr>
<td>Macrocytosis</td>
<td>0%</td>
</tr>
<tr>
<td>Spherocytosis</td>
<td>6%</td>
</tr>
<tr>
<td>Anisocytosis</td>
<td>25%</td>
</tr>
<tr>
<td>Poikilocytosis</td>
<td>16%</td>
</tr>
<tr>
<td>Polychromasia</td>
<td>8%</td>
</tr>
<tr>
<td>Fragmented cells</td>
<td>12%</td>
</tr>
</tbody>
</table>
58% of patients had anemia with hemoglobin levels of less than 12.0gm/dl and about 7% had severe anemia with hemoglobin levels of less than 8gm/dl. The incidence of anemia has been reported to be 4-25% in different studies done previously. The etiology of anemia in malaria is multifactorial. Intravascular hemolysis, removal of infected cells by spleen, immune complex adsorption onto erythrocyte membranes, therapeutic agents action on parasitized cells and bone marrow erythroid hypoplasias are the possible causes of anemia in malaria.

Majority of the patients (82%) in this study had normal leucocyte counts. Leucopenia was seen in 3% of the patients only while 15% had normal leucocyte counts. Leucopenia was seen in therapeutic agents action on parasitized cells and various ways with anemia and thrombocytopenia showed leukopenia, which was contrary to results leukocytosis. In our study, only 3% patients were lower than those in uninfected patients. Those having an infection with falciparum were relatively higher in our study. Leukocytosis may be suggestive of co-existing viral infections particularly in the presence of atypical lymphocytes common in children with concurrent viral infections. Adedapo et al reported leukocytosis among 9.5% of patients of malaria.

70% of the patients with malaria had thrombocytopenia in this study. This observation was comparable to earlier studies reporting thrombocytopenia. Memon has reported the incidence of thrombocytopenia in malaria to be 70%. In the study done by Nadeem et al thrombocytopenia was observed in 83% of P. falciparum patients and 70% of P. vivax patients.

Although the exact mechanism of thrombocytopenia in P. vivax is still a topic of extensive worldwide research, some of the postulated mechanisms are immunoglobulin G (IgG)-mediated platelet destruction, sequestration in the spleen, oxidative stress, and abnormalities in platelets’ structure caused by the invasion of the parasite as possible explanation. Researchers have also suggested thrombocytopenia as a result of consumption by disseminated intravascular coagulation (DIC) and peripheral platelet destruction induced by P. falciparum, although the latter mechanism has not been systematically evaluated in P. vivax malaria.

CONCLUSION
Plasmodium vivax typically affects blood indices in various ways with anemia and thrombocytopenia being the frequent most hematological outcomes. Hypochromia and microcytosis were the common most microscopic findings in such patients.

REFERENCES


