

ORIGINAL ARTICLE

Blood Morphology of Patients Suffering from MalariaMohammad Sajjad¹, Zard Ali Khan², Mohammad Akram³, Mokamil Shah⁴**ABSTRACT****Objective:** To find out changes in peripheral blood of patients suffering from malaria.**Study Design:** Descriptive case series.**Place and Duration of Study:** The study was conducted at Shah Noor Medical Laboratory, Bannu Khyber Pakhtoon khawa from 1st May 2014 to 31st August 2014.**Materials and Methods:** In this study 160 cases of malaria patients were selected by non probability random sampling technique with informed consent of the patients. These patients were subjected to complete blood counts performed by Hematology analyzer Sysmex 21 and manual method using Neubaur's chamber and thick and thin films stained with Giemsa stain to confirm malarial parasite. The data was recorded in preformed designed proforma and was analysed by using a Statistical Package for Social Sciences (SPSS) version 16 for percentages, means and standered deviation.**Results:** Out of 160 patients 93(58%) were males and 67(41.87%) females. Most 59 (36.8%) were in the age range of 1-10 years. Plasmodium vivax found in 135(84.3%) and Plasmodium falciparum in 25(15.6%) cases. Minimum total leukocyte count (TLC) was 1300/cmm and maximum was 19500/cmm, Low Hemoglobin level was 4.50 g/dl and high value was 15.20 g/dl. Platelets minimum count was 35000/cmm and maximum was 590000/cmm. Anemia was present in 103(64.37%), thrombocytopenia was present in 65 (40.62%), leukopenia was present in 34 (21.25%), bicytopenia was present in 24 (15%) and Pancytopenia was found in 5 (3.12%) of cases, where as high TLC was encountered in 6 (3.75%) cases.**Conclusion:** This study shows that person suffering from malaria have significant changes in blood morphology especially cytopenias causing morbidity and mrtality needs special attention regarding accurate diagnosis and prompt treatment to avoid the complications.**Key Words:** *Malaria, Plasmodium Vivax, Blood Smear, Thrombocytopenia, Pancytopenia.***Introduction**

Malaria is the most common parasitic disease transmitted by an infected female anopheles mosquito. Malaria transmission occurs in all six WHO regions. Globally an estimated 3.2 billion people are at risk of being infected with malaria and developing disease, and 1.2 billion are at high risk (>1 in 1000 chance of getting malaria in a year).¹

Four species of Plasmodium can cause human disease, Plasmodium falciparum, vivax, ovale and malariae. The disease is endemic in tropical and subtropical areas of Asia, Africa and North and South America. Worldwide most of the malarial infections are caused by plasmodium vivax, it is the most common infection in Pakistan especially in Khyberpakhtoonkhwa.² Pakistan in the group 3

countries of Eastern Mediterraeen region (Afghanistan, Djibouti, Sudan, Somalia and Yemen) shares about 95% of the total burden.^{3,4}

Malaria if untreated may cause severe morbidity and mortality. Major factors responsible in malaria for morbidity and mortality are anemia and cytopenias. Thrombocytopenia is a well-known complication of malaria and has been present in significant number of malaria patients. The white blood cell count in malaria is usually normal but it may be low or raised in severe cases.⁵⁻⁸

Hematological changes in malaria may vary with severity of infection, endemicity of malaria, hemoglobinopathies, nutritional status of patient, demographic factors, and patient immune status. Hematological manifestation of malaria are mainly due to the infection of red blood cells (RBC) by the parasite. The ability of the various plasmodia to infect RBCs is related to their attachment to specific red cell membrane receptors e.g plasmodium vivax and ovale invade only reticulocytes, plasmodium malariae invades only mature RBCs and plasmodium falciparum invades erythrocytes of all ages, as a result the proportion of RBCs parasitized in vivax

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rarely exceeds 1 % and in contrast in falciparum as many as 50% of cells may be infected indicating that various plasmodium species use different receptor.^{1,9-14}

The incubation period for plasmodium vivax is usually between 12 and 17 days, but it may be prolonged to 8-9 months or even longer, in plasmodium falciparum and ovale infection this period is from 12 to 18 days and it forms liver hypnozoites and in plasmodium malariae it is 18-40 days; it does not form liver hypnozoites.^{15,16} Malaria is diagnosed by microscopic examination of the blood thick and thin films from finger prick after aseptic measures. Two drops of blood are placed on the slide, one for thin smear and other for thick smear. For making good smears practice is mandatory.

Monocytes containing pigment are cleared more slowly than pigment containing neutrophils.¹⁷ Before a thick film smear is judged to be negative, 100 to 200 fields should be examined under oil immersion. Phagocytosed malarial pigment is sometime seen inside peripheral blood monocytes or polymorphonuclear leukocytes and may provide a clue to recent infection if malaria parasites are not detected.¹⁸

The diagnosis of malaria rests on the demonstration of asexual forms of parasite in the stained peripheral blood smears. After a negative blood smear, repeat smears should be made if there is a high degree of suspicion.¹⁹

Chinese researchers have shown that smears from intradermal blood may contain more mature forms of plasmodium falciparum than the peripheral blood. This is considered to allow a more complete assessment of severe malaria. The smears are taken from multiple intradermal punctures with a 25 gauge needle on the volar surface of the upper forearm. The puncture should not ooze blood spontaneously, but sero-sanguinous fluid can be expressed on the slide by squeezing.²⁰

Changes in Peripheral Blood: Most commonly observed changes in the peripheral blood of malarial patients are thrombocytopenia, anemia and changes in leukocytes.

Anemia: Hemoglobin less than 10.0 g/dl

Thrombocytopenia: Platelet count less than 150,000/cmm

Leukopenia: Total leukocyte count less than 4000/cmm

Leukocytosis: Total leukocyte count more than 11000/cmm

Materials and Methods

This was a descriptive case series study performed in Shah Noor Medical Laboratory at Bannu, KPK, Pakistan. The duration was three months from 1st May, 2014 to 31st August, 2014. The inclusion criteria was all those patients with diagnosed malarial infection on blood smears. Exclusion criteria was malaria patients suffering from known chronic illnesses like tuberculosis or malignancy. In this study 160 cases of malaria patients were selected by non probability random sampling technique with informed consent of the patients. All the malaria positive patients who came to the Shah Noor Medical laboratory for malaria test were selected for this study. Two ml blood was taken using 5 cc disposable syringe, slide both thick and thin were prepared and rest of the blood was placed in EDTA tube for hemoglobin estimation, total leukocyte count and platelets count by both hematology analyzer sysmax KX 21 and were also confirmed manually.

The data of positive malarial cases and laboratory findings of complete blood count investigations were recorded on a proforma specially designed for this purpose.

All the studied variables like age, sex, type of parasite, hemoglobin level, total leukocyte count and platelets count were analyzed for descriptive statistics like percentages, mean and standard deviation as well as male to female ratio was calculated by using a computer program Statistical Package for Social Sciences (SPSS) version 16.

Results

In this study it was found that out of 160 malaria patients 93 (58.2%) were males and 67 (41.8%) were females, with male to female ratio of 1.38:1. Most common age group of in this study was from 1-10 years with the frequency of 59 (36.8%) cases, 39 (24.37%) patients were in the age group of 11-20 years, 27 (16.87%) cases were in the age group of 21-30 years, 19 (11.87%) cases were in the age group of 31-40 years and 16 (10%) cases were in the age group of 41 years and above. Minimum age in this study was 02 year and maximum was 65 years. (Table I).

Amongst the various malarial parasites plasmodium vivax was present in 135 (84.33%) cases and plasmodium falciparum was found in 25 (15.67%) cases, where is no case of plasmodium ovale and malaria were identified. (Table II).

The normal hemoglobin (Hemoglobin level more than 10 gm %) was present in 110(68.75%) of cases. Anemia (Hemoglobin level less than 10 gm %) was present in 50(31.25%) of cases. The lowest hemoglobin level was 4.50 gm% and maximum was 15.20 g%, mean was 11.43 g/%.

Regarding peripheral blood picture it was found that minimum total leukocyte count (TLC) was 1300/cmm and maximum was 19500/cmm, mean was 7436.25/cmm. Low count was found in 34 (21.25%) of cases and high in 6(3.75%) of cases where is it was normal in 120 (75%) of cases.

The normal platelet count is 150,000 to 450,000/cmm. The lowest platelets count noted was 35,000/cmm, where is highest platelets count was 590,000/cmm. In total of 160 patients 94 (58.75%) patients platelets count was normal, in 64(40.62%) it was low and in 1(0.62%) it was high i.e reactive thrombocytosis was present. Bicytopenia was present in 24 (15%) and pancytopenia was observed in 3(1.87%) cases of malaria patients in this study. (Table III).

Table I: Age group and gender distribution of malarial patients (N=160)

Age group	No of male patients/%age	No of female patients/%age	Combined no./%age
1-10	31(19.37%)	28(17.5%)	59(36.8 %)
11-20	25(15.62%)	14(8.75%)	39(24.37%)
21-30	19(11.87%)	08(5.00%)	27(16.87%)
31-40	10(6.25%)	09(5.62%)	19(11.87%)
>40	8(5.00%)	8(5.00%)	16(10.0%)
Total	93(58.12%)	67(41.87%)	100 %

Table II: Percentage of patients suffering from types of malarial parasites (N=160)

Type of parasite	No of cases	Percentage
Plasmodium vivax	135	84.33%
Plasmodium falciparum	25	15.67%
Total	160	100%

Table III: Common hematological parameters in malarial patients (N=160)

S.no	Parameter	Normal cases (%)	Low cases (%)	High cases (%)
1	Hemoglobin	110(68.75%)	50(31.25%)	00(0%)
2	TLC	120(75%)	34(21.25%)	06(3.75%)
3	Platelets count	94(58.75%)	65(40.62%)	01(0.62%)

Discussion

Malaria is caused by a parasite called Plasmodium which is transmitted via the bites of infected mosquitoes. In the human body, the parasites multiply in the liver, and then infect red blood cells.¹ About 3.4 billion people- half of the world population are at risk of malaria. In 2012 there were about 207 million malaria cases and an estimated 627,000 malaria deaths.^{1,21}

In this study malaria was commonly found in children age group (1-10 years) 59(36.8%) followed by age group (21-30 years) and (31-40 years) 39(24.37%) and 27(16.87%) respectively. The same age group in descending order with different percentages were present in a study conducted by Yasinzi et al.¹¹ In the present study male patients suffered from malaria were 58.2% and females 41.8% with male female ratio of 1.38:1, where is in Atif et al³, and Idris et al²² males were 67.35% and 55.17% and females were 32.7%,36.1% and 44.82% respectively. Male predominance is also observed in another study conducted by Yasinzi et al¹⁶, this was 86.2% in males and 13.17% in females. Males are more susceptible to many protozoan infections than females and field and laboratory studies link this increased susceptibility to infection with circulating steroid hormones. Among humans, although the incidence of infection is often similar between the sexes, sex differences in the intensity of infection are reported in which men have higher parasitemia than women (WHO).¹

In our study of 160 cases of malaria patients, vivax malaria was found in 135(84.33%) cases and falciparum malaria was found in 25(15.67%) of cases. In Idris et al²² vivax malaria was found in 72.4%, falciparum in 24.1% and in 3.44% mixed infection was found, where is no mixed infection was found in our study. These findings are consistent with other study conducted in Karachi by Mehmood et al¹⁴, while this incidence is quite different in studies conducted in Nawabshah by Akhund et al¹⁵ and in Quetta by Sheikh et al¹⁹ show an increase incidence of plasmodium falciparum 55.55% and 65.82% respectively, and that of plasmodium vivax 44.44% and 34.17% respectively.

Malaria is an annual killer of over one million people globally and its essential co-morbidity is anemia. Anemia due to malaria is a major health problem in

endemic areas particularly for young children and in women of reproductive age (WHO).¹

In our study anemia (Hemoglobin less than 10 gm/dl) was found in 60 (37.5%) patients. Severe anemia (Hemoglobin <4.5gm/dl) was found in two cases only.

In study conducted by Aatif et al³ anemia was found in 21.5% of cases. In a study conducted by Jain et al⁸ anemia was recorded in 66 (94.28%) patients, in which 37 (56.06%) were plasmodium falciparum, 21 (31.81%) were plasmodium vivax and 8 (12.12%) had mixed plasmodium falciparum and vivax infection. In another study conducted by Abro et al²⁰ in the Infectious Diseases Unit of Rashid Hospital Dubai anemia was observed in 64% of the patients.

In our study thrombocytopenia (platelet count less than 150,000/cmm) was present in 65 (40.62%) of the cases. In a study conducted by Aatif et al³ 100% of the patients were having platelets count less than 150,000/cmm, where is 66.61% were having platelets count less than 50,000/cmm. In a study conducted by Mehmood et al⁶ at Pakistan field level II Hospital, United Nations Mission in Liberia, it was found that out of 145 diagnosed cases of malaria 109 (75.18%) had thrombocytopenia. In another study conducted by Rajput et al²¹ in India 88.88% patients with acute plasmodium vivax malaria were suffering from thrombocytopenia. Thrombocytopenia was also found in 83% of cases in a study conducted Dubai by Abro et al.²⁰ This present study results are quite different from other studies results regarding thrombocytopenia conducted locally and abroad, the reason may be either low sensitivity of automatic hematology analyzer regarding platelets count as seen in routine practice. Leukopenia was found in 34 (21.25%) of the patients, where is leukocytosis was found in 3.75% of cases. In a study conducted by Aatif et al³ leukopenia was found in 43.9%, another study conducted in India by Rajput et al²¹ leukopenia was observed in 14% and leucocytosis in 4.9% cases. Thus our study in term of leukocyte count is relatively near to these studies.

Conclusion

This study show that person suffering from malaria have significant changes in blood morphology especially cytopenias causing morbidity and mrtality needs special attention regarding accurate diagnosis

and prompt treatment to avoid the complications.

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