Original Research Article

A Cross-sectional Study to Evaluate the Seasonal Trends and Changes in Haematological Parameters in Malaria Cases in Last Five Years

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Abstract

Introduction: Introduction: Malaria is a potentially life-threatening disease in humans caused by Plasmodium. P. falciparum has major contribution in malaria related death while other species P.vivax, P.ovale and P.malariae generally cause a milder disease. Malaria cases show seasonal variation with highest cases during rainy season. In malaria hematological changes like anaemia, thrombocytopenia, atypical lymphocytosis and less commonly disseminated intravascular coagulation are evident and they also play a pivotal role in serious complications.

Aims and Objectives: Aim of this study was to determine the seasonal trends of malaria, and demonstrate the changes in hematological parameters like haemoglobin level (Hb), platelet count, Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH), Mean Corpuscular Haemoglobin Concentration (MCHC) and Red cell distribution width (RDW) in malaria patients during the five year study period in tertiary care hospital. **Materials And Methods:** This retrospective cross sectional study was conducted for a period of five years from January 2016 to December 2020 at central diagnostic laboratory of tertiary care hospital, Bareilly, U.P. Malaria was confirmed by identifying malaria parasites in thick & thin peripheral smears and by rapid antigenic test for malaria. The values of different hematological parameters of malaria patients were collected from medical records of the hospital. **Results:** A total of 635 malaria patients with 358 (56.38%) male and 277(43.62%) female were included in our study. The prevalence of P. vivax malaria was highest with 475(74.80%) cases. A total of 470(74.01%) malaria cases were observed from July to October with highest prevalence in the month of September (31.18%). From study population anemia and thromboctypenia was observed in 566(89.13%) patients and 457(71.97%) patients respectively. MCV was subnormal in 229(36.06%) patients. Low MCH was observed in 160(25.19%) patients and low MCHC in 109(17.16%) patients. RDW was raised in 246(38.74%) patients.**Conclusion:** This study concludes that infection of P. falciparum and P.vivax can cause significant changes in hematological parameters. That's why these parameters can be used as a reliable diagnostic marker for supporting malaria in presence of characteristic clinical findings and negative PBS for malaria parasite.

Keywords: Malaria, Seasonal Trend, Anaemia, Haematological Changes.

Introduction

Malaria is a potentially life-threatening mosquito-borne infectious disease in humans caused by Plasmodium. This disease is most commonly transmitted by infected female Anopheles mosquito which usually bites between dusk and dawn. The 5 species of Plasmodium that can cause infection are P.falciparum (causes most of the deaths), P.vivax, P.ovale, P.malariae (these generally cause a milder form of malaria) and P.knowlesi (rarely causes disease in humans).Climatic factors greatly influence the pattern and level of malaria transmission. Most important climatic factors that directly affect malaria transmission are temperature, rainfall and humidity. Peak of transmission is usually during the rainy season or soon after. Optimum temperature for malaria transmission ranges from 20-30℃ and humidity of >60% has a positive impact on the incidence of malaria.Approximately 2.48 million malarial cases are reported annually from south Asia, of which 75% cases are contributed by India alone[1-4]

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According to National Vector Borne Disease Control Programme (NVBDCP) of Government of India, in 2019 and 2020 total malaria cases in India were 0.34 and 0.18 million respectively. Among these cases, 46.36% and 63.32% were caused by P. falciparum in the year 2019 and 2020 respectively. Annual Parasite Incidence (API) rate has consistently come down in the past 20 years from 2.12 in 2001 to 0.25 in 2019. API was 0.13 in year 2020.[5] Although incidence of malaria has decreased in recent years according to NVBDCP still it is a major public health problem due to drug resistant malaria[5].Malaria diagnosis is usually made by microscopic examination of blood films, or with antigen-based rapid diagnostic tests. It has a good prognosis if diagnosed early and treated appropriately.Changes in hematological parameters are common in malaria patients and they play a pivotal role in development of serious complications. These include anaemia, thrombocytopenia, atypical lymphocytosis and less commonly disseminated intravascular coagulation. Leucopenia, leucocytosis, neutropenia, neutrophilia, eosinophilia and monocytosis also have been seen. Thrombocytopenia and anaemia may be due to haemolysis, decreased cell deformity of parasitized cells, increased splenic uptake, and decreased survival and production of platelets. Thrombocytopenia has been reported in majority of malaria studies[8-13]. and various immunopathological studies have shown the importance of platelets in innate immune responses against malaria infection. A retrospective study including 614 patients showed that multi-organ dysfunction was found in patients with thrombocytopenia[14-16] So understanding the

importance and significance of thrombocytopenia in malaria may help us guiding treatment accordingly. Our study objective was to describe the seasonal trends of malaria cases and assessment of changes in hematological parameters in malaria patients at a tertiary care centre in northern part of India.

Materials and Methods

This retrospective cross-sectional study was carried out at central diagnostic laboratory of a tertiary care centre of northern India. Institutional clearance was obtained from ethical committee of the medical college. The study was conducted on collected data of malaria cases taken from the medical records of a period of five years from January 2016 to December 2020. Both outpatient and inpatient cases were taken into consideration for enrolment in this study. Malaria was confirmed by identifying malaria parasites in thick & thin peripheral smears after staining them by Leishman stain and by rapid antigenic test for malaria. All peripheral smear/rapid antigen test positive patient were included in this study. Coexistence of cases of dengue, typhoid, bleeding disorder, chronic liver diseases, thrombocytopenia, drug intake or conditions which might have contributed to blood changes were excluded from the study based on clinical investigations. Of all the investigations done on the blood samples, investigations taken into consideration were total erythrocyte count, total leukocyte count, platelet count, haemoglobin level (Hb), Mean Corpuscular Volume (MCV), Mean Corpuscular Haemoglobin (MCH), Mean Corpuscular Haemoglobin Concentration (MCHC) and RDW.

Results

A total of 635 patients with confirmed diagnosis of malaria were included in our study. Out of these 358 (56.38%) were male and 277(43.62%) were female in our study (Table-1).

P. vivax malaria was more common 475(74.80%) as compared to P. falciparum cases 146(22.99%), while only 15(2.20%) cases of mixed infection were reported. (Table-2)After analyzing the data, it was found that maximum number of malaria cases occurred during monsoon season. A total of 470(74.01%) malaria cases were observed from July to October with highest prevalence in the month of September (31.18%).(Table-3)(Figure-1)Mean age of study population was 38.08±14.62 years. Mean values of hemoglobin and platelet were 9.68±1.78 gm/dl and 1.41±0.83/microl respectively. After analyzing the red cell indices like MCV, MCH, MCHC and RDW mean values were 82.84±8.80 fl, 28.28±1.77pg, 32.03±2.00 gm/dl and 15.65±2.43 respectively.(Table-4)Among study population anaemia was observed in 566(89.13%) patients. A total 457(71.97%) patients were found to have thrombocytopenia. MCV was subnormal in 229(36.06%) patients. Low MCH was observed in 160(25.19%) patients and low MCHC in 109(17.16%) patients. RDW was raised in 246(38.74%) patients.(Table-5)

Table 1: Gender wise distribution of malaria cases in different years

Years	Males	Females	Total
2016	81	43	124
2017	85	54	139
2018	92	59	151
2019	100	64	164
2020	41	16	57
Total	399(62.83%)	236(37.16%)	635

Table 2: Showing frequencies of different type of Plasmodium infection

Year	P. vivax		P. falciparum		Mixed infection		Total
	Male	Female	Male	Female	Male	Female	
2016	62	32	18	10	1	1	124
2017	66	42	17	11	2	1	139
2018	70	46	20	12	2	1	151
2019	74	46	23	16	3	2	164
2020	31	13	9	3	1	0	57
Total	303	179	87	52	9	5	635
	482(75.90%)		139(21.89%)		14(2.20%)		

Table 3: Frequency distribution of malaria cases in different months

Months	2016	2017	2018	2019	2020	Total
January	1	1	1	1	1	5(0.79%)
February	1	1	2	1	2	7(1.1%)
March	2	3	2	3	2	12(1.89%)
April	3	3	4	4	0	14(2.2%)
May	3	3	5	6	0	17(2.68%)
June	5	6	7	9	1	28(4.4%)
July	9	12	14	15	2	52(8.19%)
August	17	19	22	23	5	86(13.56%)
September	43	41	46	50	18	198(31.18%)
October	25	33	30	31	15	134(21.10%)
November	14	15	16	18	9	72(11.34%)
December	1	2	2	3	2	10(1.57%)
	124	139	151	164	57	635

Table 4: Showing demographic and hematological parameters

	Mean±SD
Age	38.08±14.62
Hemoglobin	9.68±1.78
Platelets	1.41±0.83

MCV	82.84±8.80
MCH	28.28±1.77
MCHC	32.03±2.00
RDW	15.65±2.43

Table 5: Prevalence of different abnormal hematological findings

	Prevalence
Anemia	566(89.13%)
Thrombocytopenia	457(71.97%)
Low MCV	229(36.06%)
Low MCH	160(25.19%)
Low MCHC	109(17.16%)
Raised RDW	246(38.74%)

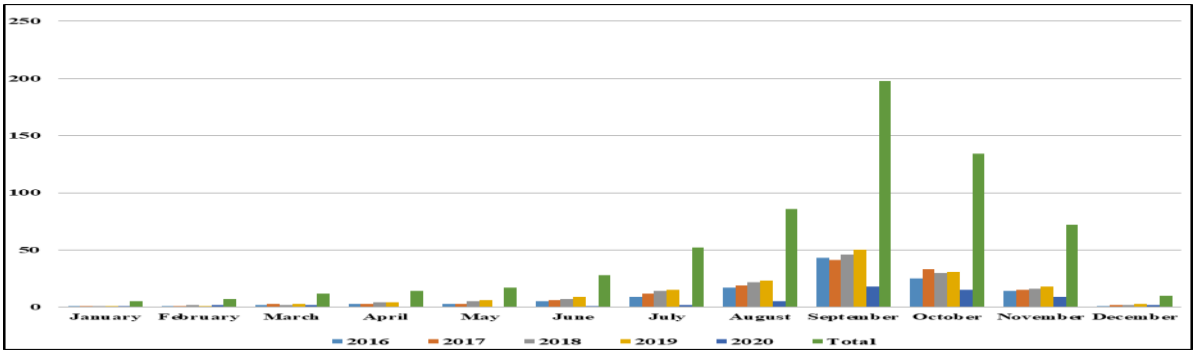

Fig. 1: Frequency distribution of malaria cases in different months

Discussion

Malaria is a prevalent disease in tropical & subtropical areas causing significant burden on health care system. It is a curable disease but delay in diagnosis and treatment increase the morbidity & mortality in patients. In malaria changes in hematological parameters are quite common although these changes are related to species of malaria. Other factors like presence of haemoglobinopathy, geographic pattern and immunity against malaria parasite also influence the hematological parameters in malaria patients.Incidence of malaria is influenced by environmental factors and poor surveillance[17-21]. Favorable environmental factors for spreading malaria are good rainfall, relative humidity of 60% and temperature between 20 and 30°C.[3] Rainfall influences the transmission of malaria by creating breeding sites and also increases relative humidity which is favorable for parasite development and disease transmission. Maximum rainfall occurs from July to September month and it has been observed that from July to October month higher number of malaria cases is recorded[22-24] According to World malaria report 2013, moderate rainfall was found to be a more favorable factor for higher malaria incidence instead of heavy rainfall. Presence of permanent vector habitation such as slow-flowing rivers and lakes provide suitable breeding sites for malaria vectors throughout the year and assist in occurrence of new cases in seasons other than rainy season.

In India, prevalence of P. vivax infection is highest about 65-70%, followed by infection of P. falciparum which is about 25-30%. Proportion of mixed infection and P. malariae is 4-8% and 1% respectively.[25] In our study the number of P. vivax malaria cases were 74.80% followed by 22.99% and 2.20% cases of P. falciparum and mixed infection respectively. Observation of our study is concordant with studies done by Upadhyayula et al[22]and Chery et al[26] Sharma et al also found similar type of result in their study[27] In our study males were more affected than females, that was comparable to study findings by Kumar et al., Karlekar et al. and Pathak et al[28-30]

Pathogenesis of anaemia in malaria cases is multifactorial. Direct parasitization of red blood cells by Plasmodium species lead to destruction of red blood cells. Accelerated removal of parasitized and non-parasitized red blood cells in spleen, suppression of erythropoietin in few cases of *Plasmodium falciparum* and anaemia of chronic disease are other important causes for that.Inflammatory mediators like Tumor necrosis factor alpha (TNF-α) may cause ineffective erythropoiesis. Prevalence of anaemia in our study is comparable with the study conducted by Tanomsri Srichaikul et al.

Mechanism of thrombocytopenia in malaria is not well understood but different studies suggest few possible mechanisms. These are-decreased thrombopoiesis with normal or increased megakaryocytes in bone marrow examination; increased sequestration of injured platelets by macrophages in the spleen in cases of *P. falciparum* malaria[35] and few studies suggest that Disseminated Intravascular Coagulation (DIC) is an important mechanism of thrombocytopenia but development of DIC is rare in malaria.In acute infection of malaria concentration of platelet specific proteins and cytokines such as beta thromboglobulin (ßTG), platelet factor 4, thromboxane A2 and prostacyclin increases and platelets become hypersensitive.Different studies suggest that increased activity of platelets reduce the possibility of bleeding in malaria patients despite significant thrombocytopenia[31-37]Changes in red cell indices like MCV, MCH, MCHC and RDW also varies significantly in malaria patients. As per study findings by Bharti Arora et al, red cell indices reduction in mean corpuscular volume (MCV) in 29% cases, mean corpuscular haemoglobin (MCH) in 30.5% cases, and mean corpuscular haemoglobin concentration (MCHC) in 20.5% cases with mean of 77.8fl, 28.4pg and 31.2 gm/dl respectively were recorded[38]Findings of our study are in alignment with this study. Similarly, RDW is increased in 38.74% of malaria patients in our study and this finding is similar to with previous study conducted in India.[39]

Conclusion

This study concludes that infection of *Plasmodium falciparum* and *Plasmodium vivax* show seasonal variations and can cause significant changes in hematological parameters. Detection of these changes by haematological investigation is relatively easy, inexpensive and technically less sophisticated. That's why these parameters can be used as a reliable diagnostic marker for supporting malaria in presence of characteristic clinical findings and negative PBS for malaria parasite.

References

1. Malaria Fact sheet N°94. WHO. 2014.
2. Caraballo H. Emergency department management of mosquito-borne illness: Malaria, dengue, and west nile virus. Emergency Medicine Practice, 2014, 16(5).
3. Bonnlander H, Rossignol AM, Rossignol PA. Malaria in Central Haiti: a hospital based retrospective study, 1982-1986 and 1988-1991. Bull Pan Amer Hlth Org. 1994; 28(1):9-16.
4. Yadav D, Chandra J, Dutta AK. Benign tertian malaria: how benign is it today? Indian J Pediatr. 2012; 79(4):525-7.
5. Ministry of Health, & Family Welfare-Government of India. (n.d.). Malaria. Gov.In. Retrieved October 14, 2021, from https://nvbdcp.gov.in/index1.php?lang=1&level=1&sublinkid=5784&lid=3689
6. Bradley D, Newbold CI, Warrell DA. Malaria. In Oxford Textbook of Medicine, eds Weatherall, D. J., Ledingham, J. G. G. & Warrell, D. A. Oxford: Oxford University Press, 1996, 835–863.
7. Butthep P, Bunyaratvej A. An unusual adhesion between red-cells and platelets in falciparum malaria. Journal of the Medical Association of Thailand 1992; 75(Suppl. 1):195–202.
8. Beale PJ, Cormack JD, Oldrey TB. Thrombocytopenia in malaria with immunoglobulin (IgM) changes. British Medical Journal. 1972; 1:345–349.
9. Pongponratn E, Riganti M, Harinasuta T, Bunnag D. Electron microscopy of the human brain in cerebral malaria. The Southeast Asian Journal of Tropical Medicine and Public Health. 1985; 16:219–227.
10. Pukrittayakamee S, White NJ, Clemens R, Chittamas S, Karges HE, Desakorn V, Looareesuwan S, Bunnag D. Activation of the coagulation cascade in falciparum malaria. Transactions of the Royal Society of Tropical Medicine and Hygiene. 1989; 83:762–766.
11. Emuchay CI, Usanga EA. Increased platelet factor 3 activity in Plasmodium falciparum malaria. East African Medical Journal. 1997; 74:527–9.
12. Lee SH, Looareesuwan S, Chan J, Wilairatana P, Vanijanonta S, Chong SM, Chong BH. Plasma macrophage colony-stimulating factor and P-selectin levels in malaria-associated thrombo-cytopenia. Thrombosis and Haemostasis. 1997; 77:289–93.
13. Erhabor O, Babatunde S, Uko KE. Some haematological parameters in plasmodial parasitized HIV-infected Nigerians. Nigerian Journal of Medicine. 2006; 15:52–55.
14. McMorran BJ, Marshall VM, De Graaf C, Drysdale KE, Shabbar M, Smyth GK et al. Platelets kill intraerythrocytic malarial parasites and mediate survival to infection. Science. 2009; 323:797–800.
15. McMorran BJ, Burgio G, Foote SJ. New insights into the protective power of platelets in malaria infection. Commun Integer Biol. 2013; 6:e23653.
16. Hanson J, Phu NH, Hasan MU, Charunwatthana P, Plewes K, Maude RJ et al. The clinical implications of thrombocytopenia in adults with severe falciparum malaria: a retrospective analysis. BMC Med. 2015; 13:97.
17. Beals PF. Anemia in malaria control: A practical approach. Ann Trop Med Parasitol. 1997; 91:713- 718.
18. Price RN, Simpson JA, Nosten F. Factors contributing to anemia after uncomplicated falciparum malaria. Am J Trop Med Hyg. 2001; 65:614-22.
19. Bouma MJ, van der Kaay HJ. Epidemic malaria in India and the El Nino Southern Oscillation. Lancet. 1994; 344:1638-39.
20. Mathur KK, Harpalani G, Kalra NL, Murthy GGK, Narasimham MVVL. Epidemic of Malaria in Barmer District (Thar Desert) of Rajasthan during 1990. Ind J Malariol. 1992; 29:1-10.
21. Sharma RC. Studies on outbreak of malaria in Muliad Village of Kheda District, Gujarat. Ind J Malariol. 1990; 27:157-62.
22. Upadhyayula SM, Mutheneni SR, Chenna S, Parasaram V, Kadiri MR. Climate Drivers on Malaria Transmission in Arunachal Pradesh, India. PLoS One. 2015; 10(3):e0119514.
23. HK Namera, VS Gohil, UV Patel, ZR Matariya Epidemiological Determinants for Malaria in Rajkot Municipal Corporation, Gujarat Healthline J. 2017; 8(1):49-54.
24. WHO. World malaria report 2013. Geneva: World Health Organization; 2013. Available from: www.who.int/iris/bitstream/10665/97008/1/978924156 4694_eng.pdf, accessed on March 11, 2014.
25. Park K. Text book of preventive and social medicine. XVII edition, 2015, 193.
26. Laura Chery, Jenifer N. Maki, Anjali Mascarenhas, Jayashri T. Walke, Pooja Gawas, Anvily Almeida et al. Demographic and clinical profiles of Plasmodium falciparum and Plasmodium vivax patients at a tertiary care centre in southmwestern India. Malar J. 2016; 15:569.
27. Sharma R, Ahmed S, Gupta S. Comparative evaluation of seasonal fevers in last 2 years at a tertiary care hospital in North India. Int J Curr Microbiol App Sci. 2014; 3(7):631-4.
28. Kumar A, Valecha N, Jain T, Dash AP. Burden of malaria in India: retrospective and prospective view. Am J Trop Med Hyg. 2007; 77(6 Suppl):69-78.
29. Karlekar SR, Deshpande MM, Andrew RJ. Prevalence of Asymptomatic Plasmodium vivax and Plasmodium falciparum Infections in Tribal Population of a Village in Gadchiroli District of Maharashtra State, India. An Int J. 2012; 4(1):42–4.
30. Pathak S, Rege M, Gogtay NJ, Aigal U, Sharma SK et al. Age-Dependent Sex Bias in Clinical Malarial Disease in Hypoendemic Regions. PLoS One. 2012; 7(4):e35592. doi:10.1371/journal. pone.0035592
31. Beals PF. Anemia in malaria control: A practical approach. Ann Trop Med Parasitol. 1997; 91:713-718.
32. Perrin LH, Mackey LJ, Miescher PA. The hematology of malaria in man. Sem Hematol. 1982; 19:70-81.
33. Tanomsri Srichaikul. Hematologic Changes in Malaria. Journal of Bangkok, Thailand, 1999.
34. Jandle JH. Hemolytic anemias caused by infection of red blood cells. In: Blood. 2nd edition. New York: Little brown and company, 1996;473-501.
35. Essien EM. Medical hypothesis: the circulating platelet in acute malaria infection. Brit J Hematol. 1989; 72:589-90.
36. Ladhani S, Lowe B, Cole AO, Kowuondo K, Newton CR. Changes in white blood cells and platelets in children with falciparum malaria: Relationship to disease outcome. Br J Haemato. 2002; 119:839-47.
37. Ali Hassan Abro, Abdullah Mahmood Ustadi, Nadeem Javeed Younis, Ahmed Saleh Abdou, Dujana Al Hamed, ed Alhaj Saleh. Malaria and Hematological changes. Pak J Med Sci. 2008; 24:287-91.
38. Sethi Bhawna, Arora Bharti, Kumar Yogesh, Aggarwal Reena. Iranian Journal of Pathology. Parasitemia and Hematological Alterations In Malaria: A Study From The Highly Affected Zones. 2013; 8(1):1-8.
39. Jairajpuri Z, Rana S, Hassan M, 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 medical journal. 2014; 29(1):12.

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