Original Article

Differentiating Between Malaria and Dengue Fever Using Hematological Parameters in an Endemic Area

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Abstract

Background

Malaria and Dengue fever are the two most common arthropod-borne diseases in tropical countries like India, and endemic areas like Mangalore. Since they share a similar clinical presentation, identifying the hematological parameters, can help differentiate between patients of malaria and dengue fever and can help with its prognosis and early treatment. This study aims to find a link between diagnostic markers that are used to discriminate between the infections, occuring in malaria-endemic areas, such as Mangalore, Karnataka.

Methods

Retrospective study was carried out at department of Pathology, Father muller medical college, Mangalore for the duration of 2 years, January 2019 to January 2021. The hematological parameters for the diagnosed cases were collected from hospital database, were analyzed and evaluated.

Results

White blood cells, neutrophils, monocytes were significantly lower in patients with DF compared to patients with malaria ($P < 0.005$). In contrast, lymphocytes, red blood cells, hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, and mean corpuscular hemoglobin concentration were significantly higher in patients with DF as compared to patients with malaria ($P < 0.005$).

Conclusion

This study concludes that hematological parameters differ in Dengue Fever and Malaria,and using these parameters- haemoglobin, PCV, RBC, total count, neutrophils, lymphocytes, monocytes, eosinophils, platelets, MCV, MCH, MCHC, age and gender, we can discriminate patients with dengue and malaria infection. In addition, using these parameters, will lead to early detection, diagnosis, and managementt of these tropical diseases.

Keywords:

Malarial parasite fluorescence test, Dengue non-structural protein antigen

Introduction

Dengue fever (DF) and malaria are the 2 most regular arthropod-borne diseases in tropical countries, especially in Southeast Asian

regions, where their endemic areas greatly overlap. [1, 2]

Dengue fever is caused by the dengue virus [DENV] in one amongst the four serotypes: DENV-1, DENV-2, DENV-3, and DENV-4.[3] According to the WHO, about 50 million patients are infected with DF annually worldwide and a couple of 2.5 billion people live in risk areas.[4]

Dengue infection may be a nonspecific febrile illness, as in DF, to a more severe bleeding tendency, thrombocytopenia, and plasma leakage [dengue viral infection, DHF].[5] Clinically, DF and other febrile illnesses share similar clinical presentation, including headache, myalgia, and rash. However, clinical presentation of DHF, bleeding and plasma leakage, are seen at a later stage, after the third or fourth day of fever. Patients are classified as having DHF, as per the WHO guidelines, having four signs: fever, thrombocytopenia [platelet count < 100 000/μL], bleeding [positive tourniquet test or spontaneous bleeding], and plasma leakage [evidence of pleural effusion, ascites or ≥ 20% hemoconcentration].[6]

The clinical spectrum of malaria is broad with manifestations such as severe anemia and respiratory distress. Most of these reports of severe and fatal vivax malaria have come from endemic regions, where the people have limited access to healthcare. [7]

In Karnataka, India, infectious disease and malaria coexists, and early differentiation between them could help to spot patients who are at high risk for DHF or severe malaria. Since the lines of treatment vary greatly, early differentiation between the 2 infections can help not only within the diagnosis but in patient prognosis still.

The objective of this study is to spot the common and unique laboratory features of both the infections, especially while studying cases from an malaria-endemic area, like Mangalore, Karnataka such as this study compared to other studies done.

Materials and Methods

A retrospective study of two-year duration from January 2019 to January 2021 on adults ages 12 and above diagnosed cases of malaria and dengue that were Identified by MPFT [malarial parasite fluorescent technique] and rapid dengue test [NS1antigen, IgG, IgM] respectively. Hematological data was collected from the hospital database.

Inclusion criteria All the adults ages 12 years and above, diagnosed as malaria and dengue fever

Exclusion criteria Subjects with comorbid conditions and deranged coagulation parameters. Subjects with other infections such as COVID 19.

Study design Retrospective Observational study

Sample size sample size of 176 cases for each group, calculated by $\eta=Z\alpha^2p[1-p]/e^2$

Statistical analysis The calculation of the parameters were presented as Mean, Standard deviation and frequency percentages after assessing the normality of the data and Test of significance t test were applied to see any significant mean differences between the two groups analyzed in Microsoft Excel.

Results

Demographic data of the study population

Out of the 352 patients who were investigated in this study maximum affected study group was between 21-30 years, 31% of study population, 35.2% being diagnosed malaria and 26.6% diagnosed dengue fever [see Table 1, figure 1]. No significant

differences relating to age were observed between the two groups [P = 0.196]. There were more women [42.2%] in the dengue group and more men [62.6%] in the malaria group. [see Table 2, figure 2].

Table 1 Age of the study population

			Dx		Total
			M	DF	
AGE	11 - 20	Count	33	22	55
		% within Dx	18.4%	12.7%	15.6%
	21 - 30	Count	63	46	109
		% within Dx	35.2%	26.6%	31.0%
	31 - 40	Count	26	33	59
		% within Dx	14.5%	19.1%	16.8%
	41 - 50	Count	18	22	40
		% within Dx	10.1%	12.7%	11.4%
	51 - 60	Count	19	24	43
		% within Dx	10.6%	13.9%	12.2%
	Above 60	Count	20	26	46
		% within Dx	11.2%	15.0%	13.1%
Total		Count	179	173	352
		% within Dx	100.0%	100.0%	100.0%

Table 2 Gender of the study population

			Dx		**Total**
			M	DF	
GENDER	F	Count	67	73	140
		% within Dx	37.4%	42.2%	39.8%
	M	Count	112	100	212
		% within Dx	62.6%	57.8%	60.2%
Total		Count	179	173	352
		% within Dx	100.0%	100.0%	100.0%

Hematological values of the study population

The following parameters were significantly lower in patients with DF as compared to patients with malaria [P-value < 0.005]: White blood cells, neutrophils, monocytes. The following parameters were significantly higher patients with DF as compared to patients with malaria [P < 0.005]: lymphocytes, red blood cells, hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, and mean corpuscular hemoglobin concentration. The following parameters were not significantly different in patients with DF compared to patients with malaria [P > 0.05]: eosinophils, basophils and platelets. [table 3, 4 and figure 1]

Discussion

In this Study, patients with DF and malaria are mostly seen in young population between 21 to 30 years, however, no significant difference between the two infections and age were observed. This was in concordance to a different study by Epelboin et al, which indicated that patients with DF are seen more in younger age group.[8] It's also been reported that the incidence of malaria in endemic areas decreases as people get older, which suggests that older age contributes to immunity against malaria.[9] One possible explanation is age-related differences in immune function resulting in differences within the balance between protective and harmful host immune responses to P. falciparum.[10] Previous studies have also suggested that the chance of developing severe dengue is larger in DENV-infected children than in adults.[11,12]

Secondly, significantly more female patients were infected in dengue group and more male patients were infected with malaria in this study, which a previous study by Carlos et al also correlated. [11] This may due to adult males reporting at least one journey into the forest or working in the forest, which may have made them more susceptible to getting bitten by a malaria-borne mosquito.

In the present study, hematological presentation was more severe and abnormalities were less frequent in patients with DF than in those with malaria. This may well be partly due to the very fact that patients with dengue presented early after the onset of symptoms. Red blood cells, Hb, Hct, MCV, MCH, and MCHC were significantly lower in patients with malaria. This could because of the parasite's primary target being RBCs, leading to an accelerated removal of both parasitized and non parasitized RBCs by the destruction of RBCs and bone marrow dysfunction. [13,14]

Table 3 Hematological values of the study population

		N	Mean	Std. Deviation	95% Confidence Interval for Mean		t test p value
					Lower Bound	Upper Bound	
HB	M	179	12.49	2.27	12.16	12.83	0.000
	D	173	13.71	1.91	13.42	13.99	
	Total	352	13.09	2.18	12.86	13.32	
N	M	179	70.07	13.39	68.09	72.04	0.000
	D	173	62.16	17.05	59.60	64.71	
	Total	352	66.18	15.78	64.52	67.83	
RBC	M	179	4.49	0.73	4.38	4.59	0.001
	D	173	4.74	0.65	4.64	4.84	
	Total	352	4.61	0.70	4.54	4.68	
PCV	M	179	37.56	6.71	36.57	38.55	0.000
	D	173	40.68	5.65	39.83	41.53	
	Total	352	39.09	6.39	38.42	39.76	
MCV	M	179	84.15	8.16	82.95	85.36	0.022
	D	173	85.98	6.56	84.99	86.96	
	Total	352	85.05	7.47	84.27	85.83	
MCH	M	179	27.90	3.10	27.44	28.36	0.002
	D	173	28.84	2.32	28.49	29.18	
	Total	352	28.36	2.78	28.07	28.65	
MCHC	M	179	33.12	1.25	32.93	33.30	0.001
	D	173	33.55	1.09	33.39	33.71	
	Total	352	33.33	1.19	33.20	33.45	

Table 4 Hematological values of the study population

Dx		N	Mean	Std. Deviation	Median	IQR		Mann Whitney test p value
						25^{th}	75^{th}	
TC	M	179	5379.33	2491.53	4900.00	3700.00	6200.00	0.005
	D	173	5006.94	2889.79	4100.00	3000.00	6300.00	
L	M	179	19.83	11.50	17.00	12.00	26.00	0.000
	D	173	29.29	17.26	25.00	17.00	41.00	
E	M	179	1.32	1.29	1.00	1.00	1.00	0.284
	D	173	1.45	1.96	1.00	1.00	1.00	
M	M	179	8.75	4.27	10.00	6.00	10.00	0.000
	D	173	7.03	3.85	8.00	4.00	10.00	
PLATELET	M	179	116581.01	77196.29	102000.00	66000.00	147000.00	0.484
	D	173	118872.83	93695.55	100000.00	44000.00	174500.00	

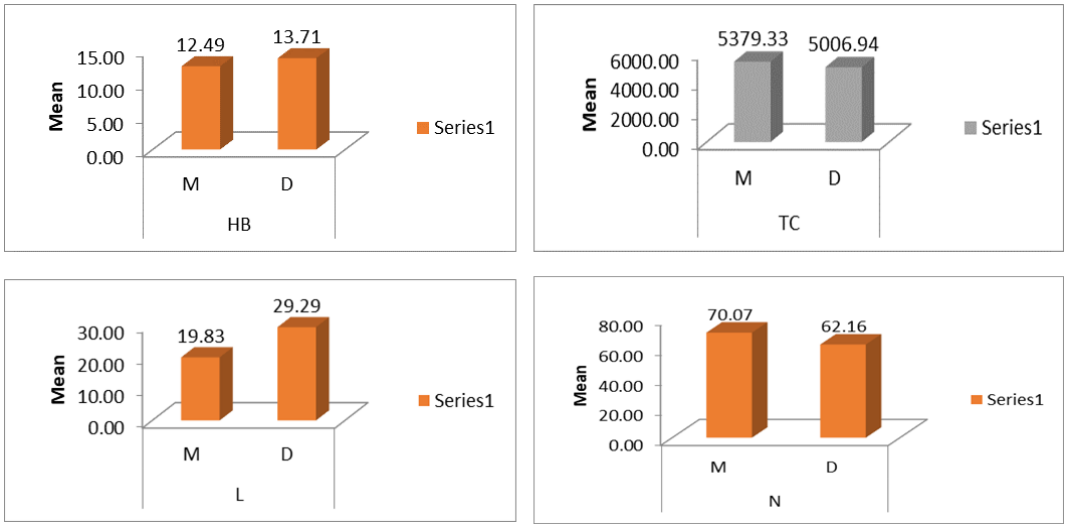

Figure 1 a. hemoglobin, b. white blood cells, c. lymphocytes, d. neutrophils

One report indicated that a high level of malaria parasitemia was related to high RBC destruction.[15] Anemia [Hb level < 11 g/dL] was frequently found in malaria positive patients, especially patients infected with the P. falciparum strain.[16] Hemoconcentration and raised Hematocrit are common in patients with DHF.[17] A high Hematocrit value is the initial abnormality occurring in DHF due to plasma leakage. Increase in Hematocrit concentration by over 20% of the baseline Hematocrit is a crucial diagnostic criterion.[18]

White blood cells, neutrophil, monocyte, eosinophil were significantly lower in patients with DF than in those with malaria. Leukopenia is common in DF patients and a useful diagnostic marker.[5] The overall decrease in leukocyte count when a patient has dengue is principally secondary to a decrease in granulocytes like neutrophils.[19] Neutropenia in dengue infections has also been reported, but less frequently,[20,21] and it had been more frequently observed during this study. A previous study by Thein et al indicated that severe neutropenia wasn't related to an increased risk of secondary bacterial infections,[22] but could also be due to leukocytes having been infected with DENV. Infected cells [dendritic cells and Langerhans cells] then migrate from the site of infection to the lymph nodes, where monocytes and macrophages are recruited, thus becoming targets of infection.[23] The suppression of WBC production in bone marrow by dengue virus is also a possible mechanism for lower leukocytes.[24] Presence of atypical lymphocytes with activated lymphocytes is reported consistently with CBC parameters,[19] but was not applicable during this study.

It has been reported that for lower eosinophil counts in patients with dengue, eosinophil concentrations fell and through convalescence, the eosinophil concentrations rose to a standard range in response to inflammation during the acute phase of the infection.[25]

A study performed in southern India by Chrispal et al to distinguish malaria to other infectious causes of fever found that normal leukocyte counts, moderate to severe thrombocytopenia, renal impairment, splenomegaly, and hyperbilirubinemia with elevated serum transaminases were related to malaria. However, rash, overt bleeding tendency, normal to low leukocyte counts, moderate to severe thrombocytopenia, and significantly elevated hepatic transaminases were related to DF.[26] A previous study by Shah et al also found that patients with DF had a high packed cell volume [PCV], whereas patients with malaria had a low PCV. Additionally, patients with malaria had low platelet count compared to patients with DF.[27] A low platelet count may be a classic

feature of both infections. This study confirmed that both patients with DF and malaria had low platelet counts, however, no significant differences between the 2 groups were observed.

This study had certain limitations like the very fact that the number of days that patients had fever before they were admitted to hospital couldn't be determined from the hospital's database. Additionally, this study lacks previous medical histories that will confound the analysis like Hemoglobin diseases, bacteria or viral infection. Further study is required to validate whether the proposed parameters were likely to be altered during the febrile stage of the illnesses.

Conclusion

The findings of this study suggest that several clinical and laboratory measures could potentially distinguish between patients with DF and those with malaria. The sensitivity, specificity, positive predictive value [PLV], negative predictive value [NPV], and diagnostic accuracy for all hematological parameters were determined. This study concludes that hematological parameters differ in Dengue Fever and Malaria and using these parameters- haemoglobin, PCV, RBC, total count, neutrophils, lymphocytes, monocytes, eosinophils, platelets, MCV, MCH, MCHC, age and gender, we can discriminate patients with dengue and malaria infection. Additionaly, using these parameters, will lead to early detection, diagnosis, and managementt of these tropical diseases, but it needs to be used in the early stages of the illnesses in order for it to be useful for reducing unnecessary medication and time of hospitalizations.

Statement of Ethics: The study protocol has been approved by Father Muller Medical College institutional Ethics and Scientific committee.

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