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Alteration of haematological parameters among patients with dengue infection.

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Abstract

Background: The objective of the present study was to evaluate the haematological parameters as screening markers for differentiating patients with dengue fever, and non-dengue fever.
Methods: This study was a retrospective case-control study in Tak Province, Thailand. All data of patients suspected with dengue infection during January 2013 and December 2015 were retrieved anonymously from computerized medical records included demographic data, medical history, and laboratory results. For statistical analysis, a normal distribution of continuous data was determined using the Kolmogorov-Smirnov test. Continuous variables were compared using the Mann-Whitney U test. Continuous variables were categorized following laboratory or usual cut-off values. Categorical variables were analyzed using Fisher's exact test or chi-square test. All analysis was performed by SPSS v.11.5 Chicago: SPSS Inc.
Results: The result showed that among 376 suspected patients with dengue infection, WBCs, neutrophils, monocytes, eosinophil's, MCV, and MCH were significantly lower in patients with DF as compared to patients with non-dengue (p value<0.05). RBCs count were significantly higher in patients with dengue infection (4.99×10^6/µL vs. 4.91×10^6/µL; p value=0.039). Platelets count were significantly lower in the patient with dengue infection compared to patients with non-dengue infection ($75{,}000 \times 10^3$/µL vs. $86{,}000 \times 10^3$/µL; p value=0.042).
Conclusion: Alteration of haematological parameters can combine with other clinical and laboratory markers which will help physicians to early diagnosis of dengue fever on the first day of admission to help closely monitoring patients with dengue and prevent developing dengue haemorrhagic fever (DHF).

Keywords: Dengue, Haematological parameters, Markers.

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Introduction

Dengue fever (DF) is caused by one of the four serotypes of the dengue virus including DEN-1, DEN-2, DEN-3, and DEN-4. This virus is an arbovirus or arthropod-borne viruses that belong to the genus Flavivirus of the family Flaviviridae [1,2]. Dengue have a variety of presentations, ranging from asymptomatic to an undifferentiated fever (dengue fever, DF) to the more severe forms such as dengue haemorrhagic fever (DHF) [3]. According to estimates of the World Health Organization (WHO), about 50 million cases of DF occur annually worldwide and 2.5 billion people live in risk areas [4].

Nowadays, laboratory techniques to confirm dengue infection are detection of viral nucleic acid and viral antigens/antibodies. The detection depends on phase of illness. At the beginning of the illness, the virus can be detected in serum, plasma, circulating blood cells and other tissues for 4-5 days. This onset of illness, virus isolation, and nucleic acid testing or antigen detection can be performed to diagnose the dengue infection [5,6]. In routine screening test of dengue, a rapid dipstick test of Dengue non-structural protein 1 antigen (NS1 Ag) Strip will serve as a useful diagnostic tool [7]. NS1 Ag, a highly conserved non-structural glycoprotein secreted by virus-infected cells, was found to increase during the acute phase of DF infection [8]. Other than antigen detection, at the end of the acute phase of infection, antibody detection is the method of choice for diagnosis [6]. However, antibody detection may take several days [9].

A previous study has showed that leukopenia is the most prominent haematological change with counts of less than 2×10^3 /µL [10]. Moreover, thrombocytopenia can occurred in 88% of the cases [10]. The decreasing platelet counts have found to predict the severity of the disease and were associated with increased haematocrit, increased liver enzymes, altered coagulation profile [11]. Early distinction between dengue and non-dengue could help clinicians to identify patients who

should be closely monitored for signs of DHF because of only patients with severe DF and DHF cases should be hospitalized. The objective of the present study was to evaluate the haematological parameters as screening markers for patients who come to the hospital for dengue diagnosis. This will help identify patients with dengue fever from non-dengue fever in order to could help the physician in better management of those patients.

Materials and Methods

The protocol of this study were followed the methods of Kotepui et al. In brief, a retrospective case-control study designed to differentiate between dengue and non-dengue was performed between January 2013 and December 2015 at the Medical Technology Laboratory of PhopPhra Hospital, Tak Province, Thailand.In brief, all data of patients suspected with dengue infection were retrieved anonymously from computerized medical records included demographic data, medical history, and laboratory results. The diagnosis of dengue was based on NS_1 Ag detection and indirect diagnosis based on the detection of specific anti-dengue immunoglobulin M (IgM) or immunoglobulin G (IgG) antibodies in patients' sera 12.EDTA blood were collected and analyzed to determine the complete blood counts (CBCs) using a BC-5200 Haematology Analyzer (Mindray, Nanshan, Shenzhen, China). Analyzer provided RBC count, haemoglobin (Hb), haematocrit (Hct), platelet count, WBC count, neutrophil, monocyte, lymphocyte and eosinophil counts, mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), and mean corpuscular haemoglobin concentration (MCHC).

For statistical analysis, a normal distribution of continuous data was determined using the Kolmogorov-Smirnov test. Continuous variables were compared using the Mann-Whitney U test. Continuous variables were categorized following laboratory or usual cut-off values. Categorical variables were analysed using the Fisher's exact test or chi-square test. Statistical significance was set at p value less than 0.05.

Results and Discussion

Demographic data of the study population

The 376 suspected patients with dengue infection were investigated (Table 1).Two hundred and thirty eight patients were confirmed with dengue infection whereas 138 patients were non-dengue infection. The rate of dengue infection among those patients was 63.3%. Median age of patients' infected and non-infected with dengue was 14 years and 15 years, respectively. Among patients infected with dengue, Thai patients (76.1%) were frequently infected with dengue than non-Thai (23.9%). No significant of age, gender, and nationality were found between the two groups (p value=0.452).

Table 1. *General characteristics.*

	Dengue	Non-dengue	P value
	n=238	n=138	(OR, 95% CI)
Demographic			
Thai/Non-Thai, n	181 (76.1)/57 (23.9)	116 (84.1)/22 (15.9)	0.066*
(%)			(0.6, 0.35-1.04)

*Comparison of 2 groups using Pearson Chi-Square, IQR: Interquartile Range; OR: Odds Ratio

Haematological values of the study population

Several haematological parameters were significantly changed in patients with dengue as compared to patients with non-dengue (Table 2). For leukocyte parameters, WBCs, neutrophil, monocyte, and eosinophil count were significantly lower in patients with DF as compared to patients with non-dengue (p value<0.01). For red blood cell (RBC) parameters, MCV and MCH were significantly lower than patients with non-dengue (p value<0.05). RBCs count were significantly higher in patients with dengue infection than patients with non-dengue infection (4.99×10^6 /μL vs. 4.91×10^6 /μL; p value=0.039). For platelets count in patient with dengue infection, it were significantly lower than patients with non-dengue infection ($75{,}000 \times 10^3$/μL vs. $86{,}000 \times 10^3$/μL; p value=0.042). However, basophils, NL ratio, ML ratio, haemoglobin, haematocrit, MCHC, and were not significantly difference in patients with dengue compared to patients with non-dengue (p value>0.05).

Table 2.*Haematological values in study population.*

Variable	Dengue	Non-dengue	P value*
	mean (IQR)	Mean (IQR)	
WBC (× 10^3/μL)	3.79 (2.64-5.31)	5.28 (3.58-7.38)	<0.0001
Neutrophil (× 10^3/μL)	2.08 (1.37-3.09)	2.90 (1.87-4.84)	<0.0001
Lymphocyte (× 10^3/μL)	1.00 (0.72-1.66)	1.33 (0.87-2.28)	0.002
Monocyte (× 10^3/μL)	0.18 (0.090.32)	0.23 (0.13-0.41)	0.006
Eosinophil (× 10^3/μL)	0.05 (0.03-0.08)	0.08 (0.04-0.14)	<0.0001
Basophil (× 10^3/μL)	0.06 (0.03-0.11)	0.08 (0.04-0.13)	0.104
NL ratio	1.71 (1.07-1.91)	2.11 (1.15-3.96)	0.108
ML ratio	0.17 (0.09-0.24)	0.18 (0.09-0.29)	0.314
RBC (× 10^6/μL)	4.99 (4.61-5.53)	4.91 (4.38-5.38)	0.039
Hemoglobin (g/dL)	13.2 (12.2-14.7)	13.2 (11.8-14.8)	0.685
Hematocrit (%)	39 (36-43)	39 (34.8-43)	0.568
MCV (fL)	80.2 (73.4-83.9)	81.3 (76.6-85.3)	0.02
MCH (pg/cell)	27.2 (24.9-28.9)	27.9 (25.9-29.3)	0.029
MCHC (g/dL)	34 (33.3-34.7)	34.2 (33.4-35.2)	0.175
RDW (%)	12.4 (12.0-13.1)	12.4 (11.9-13.1)	0.958
Platelet (× 10^3/μL)	75 (48.8-100.3)	86 (48-144.5)	0.042

*Comparison of 2 groups using The Mann-Whitney U Test

In the present study DF was more common in females (55.5%). This is in accordance with a study in Delhi, India indicated that Dengue seropositive was found to be significantly associated with the female gender [10-12]. However, general studies both sexes are equally affected although a male to female ratio of 0.65:1 [13]. Some studies found twice the number of male patients infected with dengue compared to females [14,15].

DF was more common among the paediatric age group the largest proportion was seen in the age group of 9-25 years. This was in accordance with previous reports in Thailand indicated that the age group with the highest incidence changed from those aged 5-9 years to those over 15 years of age [16-18]. This finding was consistent with the idea that the observed age shift might be a consequence of the demographic transition in Thailand.

This study revealed haematological changes in study population. WBC count was significantly lower in dengue infected patients. This is in accordance with previous studies indicated that patients with dengue had significantly lower white blood cell (WBC) counts [3,19]. This study, neutrophils and lymphocytes counts were significantly lower in DF compared to non-DF. This was in accordance with results of previous studies [3,19-22]. Beside monocytes and eosinophil count were also significantly lower in DF compared to non-DF. This is in accordance with a previous study indicated leukopenia was the most prominent haematological changed during dengue infection [10,19]. Ration of low number of white blood cell and their absolute counts may be due to the reduction in WBC due to bone marrow suppression by dengue virus [23].

This study, lower RBC count, MCV, and MCH in patients with dengue were observed. This never been reported in any previous studies. This could be explained by occurring of haemolyticanaemia in dengue infected patients. However, haemolyticanaemia in dengue fever is considered rare and has been described in case reports in Malaysia, Sri Lanka, India and in a British traveller [24-27].

This study, lower platelet count was observed in patients with dengue compared to non-dengue. This is in accordance with a previous study indicated significantly lower platelet [3]. According to the results, low platelet count is one of the criteria for diagnosis of DHF [5]. The ration behind thrombocytopenia among patients with dengue is currently unknown. However, previous studies indicated decreased production of platelets in DF and increased destruction of platelets in DHF [19,28].

The previous studies indicated that routine laboratory markers help to reduce the cost for laboratory diagnostic screening [20,29]. In routinely diagnosis of dengue infection, dengue non-structural protein 1 antigen (NS1) is an antigen presenting in sera of dengue patients during acute phase of infection and responsible for pathogenesis of dengue [30]. NS1 Ag strip has a rapid, sensitive, and easy to use for the early diagnosis of DF at the presentation [31]. Moreover, several commercial kits available on the market for detecting anti-dengue antibodies such as Immunochromatographic test (IgM/IgG detection) [32,33]. This study found that routine laboratory markers including MCV, neutrophils, MCHC, and lymphocytes are useful in detecting laboratory-confirmed dengue infection. This predictive technique could be used to decide the priority of NS1 Ag strip.

There are several limitations in this study. Firstly, this study was the retrospective and single based-hospital might not reflect the large groups of dengue patients. However, these results may provide useful routine laboratory markers for diagnosing dengue in endemic area,which may help alertness of medical technologist or medical sciences technician performing dengue diagnosis as no single laboratory marker available for predicting DF infection [34].

Conclusion

Alteration of haematological parameters can combine with other clinical and laboratory markers which will help physician to early diagnosis of dengue fever in the first day of admission to help closely monitoring patients with dengue and prevent developing dengue haemorrhagic fever (DHF).

Ethics approval and consent to participate

The retrospective use of anonymous patient files was authorized and approved by the PhopPhra Hospital and the Ethical Clearance Committee on Human Rights Related to Researches Involving Human Subjects of Walailak University. All data collected retrospectively were anonymized in a standardized case report form and in the database. The written informed consent could not be obtained from individual patients as theauthors collected retrospective data from the hospital. In this case, this manuscript, age and sex were not show to hold the anonymity of the patients.

Availability of Data and Materials

The datasets used during the current study are available from the corresponding author based on reasonable request.

Consent for Publication

Not applicable

Competing Interests

The authors declare that there is no conflict of interest regarding the publication of this article.

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Authors' Contributions

MK, KK, BP, NP, and WP participated in the study design, data analysis, and writing of the paper. All authors read and approved the final paper.

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