ISSN: 2474-3658

Simon-Oke and Akinbote. J Infect Dis Epidemiol 2020, 6:166
DOI: 10.23937/2474-3658/1510166
Volume 6 | Issue 5
Open Access

Journal of
Infectious Diseases and Epidemiology

RESEARCH ARTICLE

Prevalence of Malaria and Typhoid Coinfection in Relation to Haematological Profile of University Students in Akure, Nigeria

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Abstract

Introduction: Malaria and typhoid fever are endemic febrile diseases with overlapping signs and symptoms notably fever, diarrhoea, vomiting and headache.

Objective: The study was aimed to investigate the level of co-infection of malaria and typhoid fever and the haematological parameters among students of the Federal University of Technology Akure.

Methodology: A cross-sectional study was conducted on Two hundred students. Venous blood samples were collected for Widal and Malaria test. The Widal agglutination test was performed by the rapid slide titration method using commercial antigen suspension (Cal-Test Diagnostic Inc. Chino, U.S.A.) and Malaria diagnosis was carried out using antigen based- Rapid Diagnostic Test (RDT).

Results: Of the 200 examined students, 121 (60.5%) were positive for malaria and 161 (80.5%) were positive for typhoid fever. Male were more infected with both malaria and typhoid fever (43% and 40.5% respectively), however, the difference in the prevalence of infections between the genders were not statistically significant ($P > 0.05$). 103 (51.5%) students had co-infection out of which 78 (39%) males were more infected than 25 (12.5%) females. Age group 21-25 years had the highest co-infection (31%). There is a significant difference between the numbers of students with co-infection and those without co-infection ($P < 0.05$). In the haematological analyses, this study showed that a reasonable percentage of malaria and typhoid fever infected patients were anaemic (15.5%), 5.8% had higher than normal leucocyte count, 13.6% with lymphocyte count lower than normal and 7.8% of the co-infected students had monocyte count higher than the normal range.

Conclusion: The incidence of typhoid and malaria co-infection will greatly reduce when there is improved personal hygiene, targeted vaccination campaigns and intensive community health education on preventive and control measures against malaria and typhoid fever.

Keywords

Co-infection, Malaria, Typhoid, Plasmodium, Salmonella, Haematological parameters

Introduction

Malaria is caused by obligate intracellular parasites, which live in host erythrocytes and remodel these cells to provide optimally for their own needs. It is a major public health problem in tropical areas, and it is estimated that malaria is responsible for 1 to 3 million deaths and 300 to 500 million infections annually [1]. Malaria remains the most complex and overwhelming health problem, facing humanity in vast majority of tropical and sub-tropical regions of the world, with 300 to 500 million cases and 2 to 3 million deaths per year [2]. About 90% of all malaria deaths in the world today occur in the sub-Saharan Africa and this is because majority of infections are caused by *Plasmodium falciparum,* the most dangerous of the four human malaria parasites accounting for an estimated 1.4 to 2.6 million deaths per year in this region [3]. Typhoid fever is a bacterial infection due to *Salmonella typhi. Salmonella*'s genus is Gram-negative, motile, non-sporing, non-capsulate bacilli which exist in nature primarily as parasites of the intestinal tract of man and other animals. *Salmonella typhi* and the paratyphoid bacilli are found only in the intestinal tract of man for whom they have a high degree of pathogenicity and in which they frequently cause invasive disease that causes symptoms which may vary from mild to severe and usually begin six to thirty days after exposure with gradual onset of a

Citation: Simon-Oke IA, Akinbote MO (2020) Prevalence of Malaria and Typhoid Coinfection in Relation to Haematological Profile of University Students in Akure, Nigeria. J Infect Dis Epidemiol 6:166. doi.org/10.23937/2474-3658/1510166

Accepted: October 03, 2020: **Published:** October 05, 2020

high fever after several days [4]. Weakness, abdominal pain, constipation, and headaches are the commonest symptoms. Some people develop a skin rash with rose colored spots. Without treatment, symptoms may last for weeks or months. Other people may carry the bacterium without symptoms; however, they are still able to spread the disease to others [5].

Typhoid and malaria fever are two leading febrile illness affecting humans, especially in sub-Saharan Africa. They remain the diseases of major public health importance and the cause of morbidity and mortality. An association between malaria and typhoid fever was first described in 1862 in North America as an entity called typho-malaria fever [6]. Both diseases are common in many countries of the world where poor sanitary habit, poverty and ignorance exist. The first non-specific manifestations include fever, headache, abdominal pain and vomiting. Despite the importance of concurrent malaria and typhoid fever in the tropics, the challenges associated with the diagnosis and the public health implications have not been comprehensively reviewed. The objective of this study was to evaluate the prevalence of malaria and typhoid fever coinfection in relation to the haematological profile of the sampled population.

Materials and Methods

Study area

The study was carried out at the Federal University of Technology, Akure, Ondo-State, Nigeria. Akure is the capital and the largest city in Ondo state which covers a land area of 14,793 square kilometers within south-west of Nigeria. The Latitude of FUTA lies between 7.3043° N and longitude 5.1370° E coordinates. FUTA is estimated to have a population of about 15,000 students with 50 departments.

Sampling design

Two hundred (200) students were randomly sampled across the school for this study, irrespective of their sex, age and department. At least one student in each department was sampled.

Sample collection

Three millilitres (3 ml) of blood sample was collected from each patient into sterilized EDTA® falcon tube by trained phlebotomist.

Malaria diagnosis using antigen based- rapid diagnostic test (RDT)

The test kit (Paracheck-Pf test kit) was detached from its seal and the blood from the EDTA bottle was blotted into sample window (S) present on the test kit, two drops of Malaria parasite test kit buffer was added to the blood sample in the sample window (S) and allowed to flow through the chamber labeled test (T) and control (C) windows. The test was allowed to run for 10 mins. The appearance of coloured band line at the control window (C) only, indicates a valid but negative test. Coloured band lines appearing at the test window (T) and control window (C) indicates a positive test. If there was no redline appearing at the control window (C), it is thus interpreted as invalid and the test is repeated.

Widal test

The agglutination test was performed on all blood samples by the rapid slide titration method using Cal-Test Diagnostic Inc. Chino, U.S.A. Widal commercial antigen suspensions, for the somatic (O) and flagella (H) antigens by adding one drop of the widal antigen suspension to the reaction circles containing the patient's serum. The content of each circle was uniformly mixed over the entire circle with separate mixing sticks. The slides were gently rocked back and forth, and observed for agglutination for one minute. A positive widal test was considered for any serum sample with antibody titre ≥ 1:160 to the O and H antigens of *S. typhi*.

Estimation of packed cell volume (PCV) and haemoglobin (Hb)

The aim of the PCV test was to measure the volume of packed red cells present in the blood. The principle of the PCV test is the ability of different blood components (red and white blood cells, and platelets) to pack together according to their rate of sedimentation after centrifuging for 5 minutes using the Haematocrit centrifuge. Similarly, Haemoglobin was also estimated by Sahli's haemoglobinometer (acid haematin method).

Total white blood cell (WBC) count

The total WBC count was determined by measuring 0.38 ml of Turk's solution using 1 ml pipette into a clean cuvette in which 0.02 ml (20 µL) of blood sample was measured using a micropipette, mixed and incubated for 1 hour. WBC count was read microscopically by counting each of the cells as seen on the Hemocytometer (Neubaer's counting chamber).

WBC differential count

The identification of the different types of white blood cells was done. In identifying the numbers of different WBC, a thin blood film was made, stained with Leishman stain, observed microscopically using the X100 objective and a large number of WBC (at least 100) was counted. This gave the percentage of cells that are of each type. By multiplying the percentage by the total number of WBC, the absolute number of each type of WBC was obtained. Five types of WBC were encountered; lymphocytes, monocytes, neutrophils, eosinophils, and basophils.

Results

Socio-demographic data

A total of 200 Students were examined, out of which

Table 1: Socio-Demographic data of students.

Variables	Groups	Frequency	Percentage (%)
	Males	103	51.5
Sex	Females	97	48.5
	16-20 years	111	55.5
Age	21-25 years	87	43.5
	> 25 years	2	1

Table 3: Haematological Profile of Malaria infected Students Frequency (%).

	Frequency (%)		
Parameters	**Below Normal**	**Normal**	**Above Normal**
PCV & Hb	20 (16.5)	101 (83.5)	0 (0)
Leucocyte	4 (3.3)	113 (93.4)	4 (3.3)
Monocyte	0 (0)	111 (91.7)	10 (8.3)
Lymphocyte	18 (14.9)	101 (83.5)	2 (1.7)
Neutrophil	0 (0)	119 (98.3)	3 (2.5)
Eosinophil	0 (0)	113 (93.4)	8 (6.6)
Basophil	0 (0)	115 (95.0)	6 (4.9)

103 (51.5%) were males and 97 (48.5%) were females. The mean age was 19.61 ± 0.177 years. 111 (55.5%), 87 (43.5%) and 2 (1%) of the students were within the age range of 16-20, 21-25 and > 25 years respectively (Table 1).

Prevalence of malaria infection

Of the 200 students, 121 (60.5%) were positive to malaria parasite. Males recorded higher prevalence of 86 (43%) than females and age group 21-25 years 64 (32%) were more infected with Malaria (Table 2).

Prevalence of typhoid fever

A total of 161 (80.5%) students were positive for typhoid test; 81 (40.5%) males and 80 (40%) females. Age group 16-20 recorded the highest prevalence of 90 (45%). However, there is no significant difference between the age groups (*P > 0.05*) (Table 2).

Co-infection of malaria and typhoid fever

The prevalence of co-infection was 103 (51.5%). 78 (39%) males and 25 (12.5%) females were co-infected. In relation to age, the highest co-infection 62 (31%), was observed in age group 21-25. However, there was a significant difference in the age groups (P < 0.05) (Table 2).

Table 3 shows the haematological parameters in malaria infected students in which 20 (16.5%) had PCV/Hb lower than the normal, 101 (83.15%) had normal PCV/Hb and none of the patients had PCV above normal. 18 (14.9%) and 4 (3.3%) students had low lymphocyte and leucocyte count respectively. None of the studied subjects recorded below normal counts in neutrophil, eosinophil and basophil. 10 (8.3%) of the infected students had abnormal monocyte count.

Table 2: The Prevalence of Malaria, Typhoid fever and their co-infection in relation to the sex and age groups of students.

		MALARIA				TYPHOID				CO-INFECTION			
Variable	Group	Positive (%)	Negative (%)	X^2	*P* value	Reactive (%)	Non-Reactive (%)	X^2	*P* value	Positive (%)	Negative (%)	X^2	*P* value
SEX	MALE	86 (43%)	37 (18.5%)	0.02	0.016	81 (40.5%)	22 (11%)	0.02	0.665	78 (39%)	25 (12.5%)	0.02	0.000
	FEMALE	35 (17.5%)	42 (21%)			80 (40%)	17 (8.5%)			25 (12.5%)	72 (36%)		
AGE	16-20	57 (28.5%)	54 (27%)	0.02	0.001	90 (45%)	21 (10.5%)	0.02	0.489	50 (25%)	61 (30.5%)	0.02	0.001
	21-25	64 (32%)	23 (11.5%)			69 (34.5%)	18 (9%)			62 (31%)	25 (12.5%)		
	> 25	0 (0%)	2 (1%)			2 (1%)	0 (0%)			1 (0.5%)	1 (0.5%)		

The haematological parameters in typhoid fever positive students show that 23 (14.2%) of the 121 infected students had PCV/Hb below normal, 98 (60.9%) had normal PCV/Hb and none of them had PCV/Hb above the normal range. 16 (9.9%) subjects had very low lymphocyte count while 6 (3.7%) had lymphocyte count above normal. Basophil count was normal for 120 (74.5%) and below normal for 1 (0.6%) students. None of the sampled students had monocyte count below normal (Table 4).

Figure 1 shows the haematological parameters among malaria and typhoid fever co-infected students. 16 (15.5%) of the 103 co-infected patients had PCV/Hb lower than normal, 86 (83.5%) with normal PCV/Hb and 1 (1.0%) above normal. 14 (13.6%) co-infected students had below normal lymphocyte count, 87 (84.5%) normal and 4 (3.9%) above normal counts.

99 (96.1%) students had normal neutrophil count, 2 (1.9%) below normal and 2 (1.9%) above normal counts. Both leucocyte and monocyte recorded 95 (92.2%) normal counts, and 2 (3.6%) had neutrophil count.

Table 4: Haematological Profile of Typhoid fever in infected Students.

	Frequency (%)		
Parameters	**Below Normal**	**Normal**	**Above Normal**
PCV & Hb	23 (14.2)	98 (60.9)	0 (0)
Leucocyte	2 (1.2)	115 (71.4)	4 (2.5)
Monocyte	0 (0)	113 (70.2)	8 (4.9)
Lymphocyte	16 (9.9)	99 (61.5)	6 (3.7)
Neutrophil	2 (1.2)	116 (72.0)	3 (1.9)
Eosinophil	1 (0.6)	113 (70.2)	7 (4.3)
Basophil	1 (0.6)	120 (74.5)	0 (0)

Discussion

Malaria and typhoid is still a major public health problem in tropics. The prevalence of malaria and typhoid co-infection is high in this study. Malaria and typhoid fever are tropical diseases where poverty mal-nutrition, poor sanitary status, poor personal hygiene, poor health facilities, poor social service and low level of education are among the factors that make tropical areas disease laden [7]. A total of 60.5% of malaria infection was recorded in this study, this is lower than the result gotten from similar study conducted by [8] among the students of the Federal University of Technology Akure. Males were positive to malaria infection more than females (Table 1), which is similar to the report of [9] and [8] in Calabar and Akure respectively. However, this result did not corroborate the report of [7] and [10] who recorded a higher prevalence among female students (56.9%) than male students (42.9%) in Abakaliki and

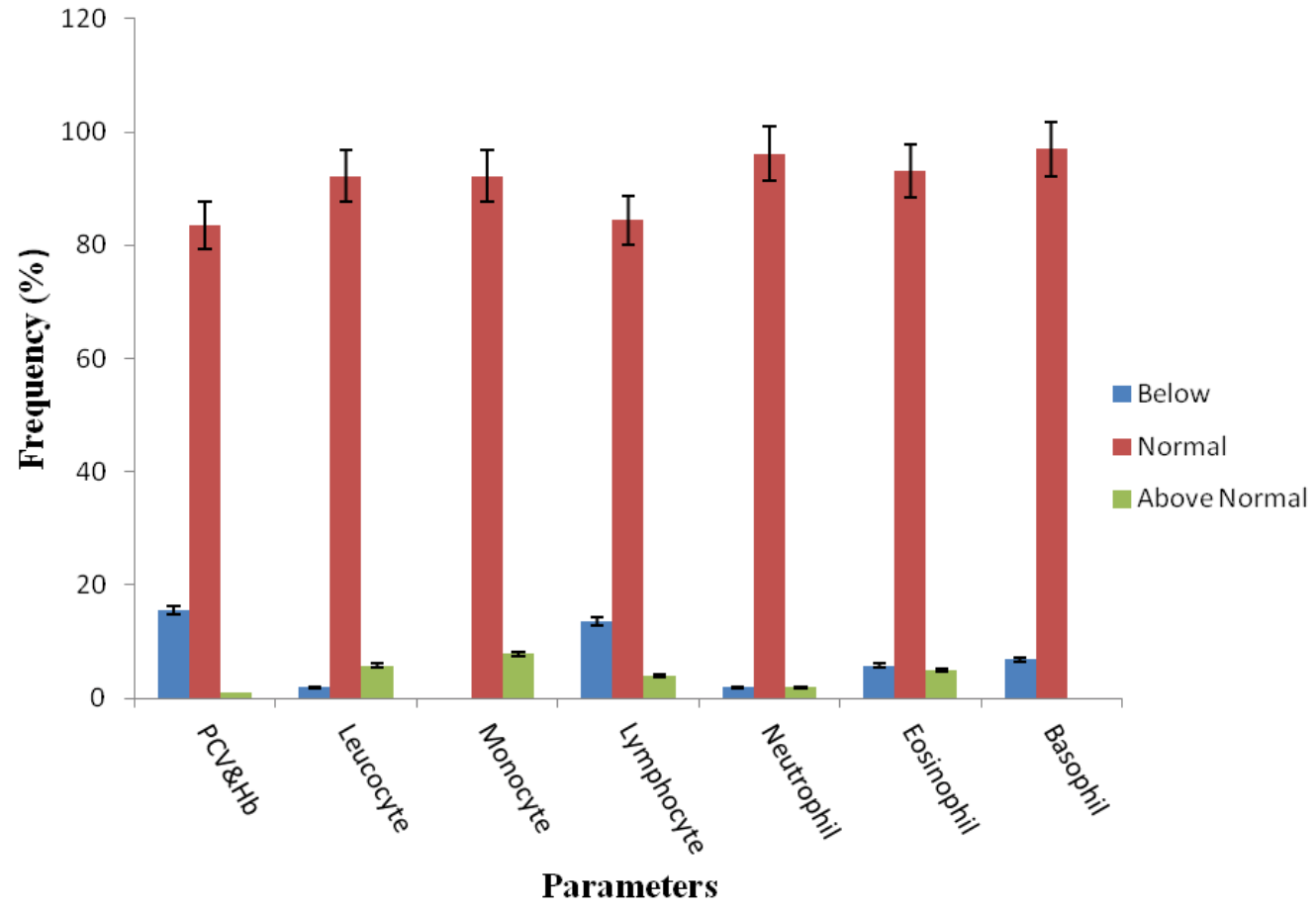

Figure 1: Haematological profiles of malaria and typhoid co-infected students.

Wukari, Nigeria. Age group 21-25 recorded the highest malaria prevalence of (52.89%). This study is in agreement with the study of [11] who recorded higher prevalence among age group 21-30 and a lower prevalence among age 11-20. The high prevalence recorded among this age group could be as a result of their different habits, level of exposure to mosquito bites and immunity developed against the parasite [12].

Typhoid fever prevalence rate was high (80.5%), slightly greater in males 81 (40.5%) than in females 80 (40.0%), but not statistically significant ($P > 0.05$). This prevalence rate is contrary to the report of [13] in Samaru, Zaria where females had slightly higher prevalence rate of typhoid fever (45.3%) than males (42.2%) but similar to the result obtained by [14] in Pakistan where the males had higher frequency of typhoid (51%) than females (49%). The high prevalence of typhoid fever could be attributed to the source of drinking water (such as water from uncovered well, contaminated sachet water) and indiscriminate defaecation in bushes around water sources which is a common practice among students which could have contributed to the high burden of typhoid fever recorded in this study [15].

The prevalence of malaria and typhoid co-infection (50.5%) in this study is higher than (10.33%) reported by [16] in Sokoto, (15.2%) by [13] in Zaria and (28%) by [9] in Calabar. However, there was a significant difference between the numbers of individuals co-infected and those not co-infected ($P < 0.05$). Males had the higher percentage of co-infection than females and age group 21-25 years-old has the highest percentage for co-infection (Table 3). This does not agree with the report of [9] in which the highest co-infection rate (37.5%) was in the age group of 1-15 years. Delay in treatment and treatment option could contribute to persistence and continued transmission of the co-infections.

This study showed notable changes in the haematological parameters of patients infected with malaria. 20 (16.5%) had haemoglobin lower than the threshold 11 mg/dL haemoglobin level which is lower than the 87.5% reported by [17] at Ahmedabad in India. However, there was no significant difference between low haemoglobin in malaria-positive patients and that in malaria negative patients ($P > 0.05$). The neutrophil and eosinophil count above normal (2.5%) and (6.6%) respectively as well as lymphocyte count below normal (14.9%) cannot be completely attributed to malaria infection alone as other complications might be involved. 10 (8.3%) of the patients had monocyte count higher than normal which correspond to the majority observation that malaria infection contributes to elevated monocyte count [10]. 23 (14.2%) of typhoid fever positive patients had lower PCV/Hb which is dissimilar to the report of [18] (34.48%) in Kalaburgi, India and [10] in Wukari, Nigeria.

Neutrophil count in typhoid positive patients was (72.0%) which agrees with the statement of [19], that neutrophil count can only be abnormally elevated in complicated typhoid fever. 16 (15.5%) of malaria and typhoid fever co-infected patients had (low PCV/Hb). Prevalence of typhoid and malaria is high in the tropics leading to co-infection. The mechanisms to explain the association between malaria and typhoid co-infection is unknown, though it has been shown that haemolysis, which occurs in malaria, may predispose to typhoid fever [20]. Lymphocyte count below normal range in co-infected students (31.6%) may be medically significant because the combined effects of the co-morbidity of malaria and typhoid fever can result in the anomaly [14]. The leucocytes count in co infected patients was higher than normal in 6 (5.8%) patients. It may be due to the body's effort to resist infection by *Plasmodium* and S*almonella* resulting in continuous production of leucocytes [21].

Conclusion

Malaria and typhoid fever co-infection was prevalent among the sampled students which is a major public health problem in many developing countries. Co-infection rate was higher in males than females and age group 21-25 years recorded the highest prevalence. Infections with malaria and typhoid fever have obvious effects on haematological parameters which could serve as useful indices during diagnosis. Since they both have similar symptoms, treatment should be based on adequate laboratory diagnosis.

Acknowledgements

The authors appreciate the assistance of the phlebotomist and the technical staff of the laboratory used for the research work.

Ethical Clearance

Prior the commencement of the research work, approval was given by the Health Research Ethics Committee (HREC) of the University, the Chief Medical Director of the University health Centre and informs consent from the sampled students who were clearly informed on the aims and objectives of the research.

Conflict of Interest

The authors declare no conflicts of interest of whatsoever.

Author's Contribution

We declare that the both authors have made substantial contribution to the concept, acquisition and interpretation of data, draft of the article and its critical revision. Both authors approved the submission of the final version.

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