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Distribution and Co-infection of Malaria and Syphilis in relation with ABO and rhesus among students of Federal University of Technology, Akure Ondo State, Nigeria

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ABSTRACT

The human ABO blood and Rhesus could be an important determining factor in the distribution and co-infection of human malaria parasite and Syphilis infection. Owing to the widespread of malaria, reckless lifestyle and attitudes of present-day youths towards hygiene and sex-life, and increasing sexually transmitted infections (STIs) amongst students, it is crucial to assess the risk of exposure, distribution, and ascertain the most susceptible blood group to these infections. Malaria is an important cause of ill-health primarily transmitted by Female Anopheles mosquito and is responsible for severe morbidity and mortality especially in Sub-Sahara Africa and globally; while coinfection is the single or collective habitation of pathogenic organism in host body. Syphilis is an infection caused by *Treponema sp* usually transmitted by sexual contact via a chancre. This work investigated the possibility of individual susceptibility to malaria and syphilis infection when their ABO blood types and Rhesus is considered a predisposing factor. Two millimetres (2ml) of venous blood was obtained by venipuncture from asymptomatic subjects into ethylene-diamine-tetra-acetic-acid (EDTA) anticoagulated blood containers, properly mixed and labelled appropriately. 229 respondents were subjected to ABO blood-typing using monoclonal Antisera A, B, AB, D and microscopic analysis for Plasmodium parasites screening using thick and thin smear approaches. Antibody specific test against treponema antigens (RDT) was also carried out using whole blood. Of the sampled individuals, 199(87%) and 30(13%) were rhesus D positive and negative respectively, where 75.1% was positive for Plasmodium infection. The relationship between ABO blood group and malaria parasite was significant in the study population ($P = 0.002$, $p < 0.05$). Furthermore, the distribution of malaria and syphilis in relation to ABO (see Fig 4)

suggests that infection is equal among gender ($X^2 = 1.767$, $P = 0.184$, $p > 0.05$) and may be at risk under any given circumstances.

Keywords: Asymptomatic Malaria, Syphilis, Co-infection, Prevalence, Sexual-health, ABO system, Youths and student's lifestyle

1. INTRODUCTION

Malaria is a mosquito-borne disease caused by Plasmodium species. It presents clinical manifestation such as fever, headache, shivering, nausea, vomiting, arthralgia, anaemia, hepatomegaly, convulsion, death and retinal whitening (Bittaye et al., 2022). Malaria is the most important parasitic disease of man, (White, 2018) which is believed to be a major obstruction to social and economic development in Africa because it causes enormous misery and suffering through the pain of fevers and the anguish of bereavement.

It is estimated that more than 300 million acute cases of malaria occur worldwide each year, resulting in one million deaths, where ninety percent (90%) of these deaths are in Sub-Saharan Africa, and most victims are children aged less than five years (Schumacher and Spinelli, 2012). Moreover, there are about 95,000 annual deaths of children less than five years old (Dasgupta et al., 2022). Malaria is implicated in several off-school days cases in affected areas where deaths are recorded daily and rob victims of realizing future goals and prospects. Nigeria is one of Africa's hardest hit countries with approximately 51 million cases and 207,000 deaths annually (Dawaski et al., 2016).

This magnitude of occurrence in this part of the world correlates with poverty, ignorance and social deprivation in the community. There are five species of Plasmodium that infects humans: *P. falciparum*, *P. vivax*, *P. ovale*, *P. malariae* and *P. knowlesi* (Sato, 2021). Several studies show that *P. falciparum* is the deadliest form of malaria especially in sub-Saharan Africa where it accounts for about 98% of the total malaria cases in Nigeria (Chijioko-Nwauche et al., 2015). Malaria imposes varying intensity of social, educational, cognitive and financial burdens which are exclusively not because it is a disease of the poor but with no constant effect across all races (Olajide, et al., 2022; Onukogou, et al., 2018).

Syphilis is a sexually transmitted genital ulcerative disease caused by *Treponema pallidum*, which is a fragile spiral bacterium majorly transmitted through an open genital wound or lesion during sexual contact because it can survive only briefly outside of the body (Spicknall et al., 2018). Syphilis is a worldwide disease which is particularly distributed in countries with low socio-economic status. Annually, there are about 6 million new cases in persons aged 15-49 years (Kojima and Klausner, 2018). The global incidence rate of syphilis was 1.5 cases per 1000 males and females respectively, where 18 million prevalent cases of syphilis were recorded in adolescents and adults aged 15-49 (Newman et al., 2015). According to the same report, the highest prevalence was in the African region, followed by the Southeast Asian and Western Pacific regions. Furthermore, the rate of reported primary and secondary syphilis cases remains highest among Blacks, with the overall rate of syphilis being recorded in Black men (Stone et al., 2018).

The blood grouping system consists of the A, B and H carbohydrate antigens, which can regulate protein activities during infection, and antibodies against these antigens.

ABO blood grouping is based on the presence or absence of A and B blood group antigens on the surface of red blood cells (RBC) derived from inherited gene (Simon-Oke *et al.*, 2016).

The distribution of the four ABO blood types, A, B, AB, and O, varies in populations throughout the world and is determined by the frequency of the three alleles of the ABO gene.

It has been the most important selective force on the human population and may determine the severity or risk of an infection.

There is a dearth of information on the co-existence of malaria and syphilis; and recent literatures addressing the impact of ABO and Rhesus on human susceptibility to both infections are grossly insufficient. It is important to reiterate the rising sexual relations ranging from oral, anal, vaginal intercourse e.t.c that can expose students to varying STIs including syphilis; more so, with malaria being a life threatening disease and having a disproportionately high burden accrued to the African region (Alegana *et al.*, 2020), it is therefore thought worthy to assess the distribution of malaria and syphilis, their co-infection in relation to ABO blood group and Rhesus among University student of FUTA, Nigeria. This will also create an invaluable window for enlightenment about more carefulness on unguarded sexual contacts among university students and improved consciousness towards prompt alertness for proper diagnosis and treatment of malaria in order to prevent upsurge of emergency malaria cases.

2. MATERIALS AND METHOS

Aim

The aim of this study is to determine the distribution of malaria and syphilis and its co-infection among the student of FUTA in relation with ABO and Rhesus factor.

Study Area

The study was conducted at the Federal University of Technology Akure, (FUTA) Ondo State. Akure is the capital and the largest city of Ondo State which covers a land area of 14,793 square kilometres within south-west of Nigeria. It lies between latitude 7°15'0"N and longitude 5°11'42"E and has a population of about 717,000. Akure, has an average temperature of 25.6 °C and relative humidity of 85%.

Ethical Clearance

Prior to the commencement of the research, relevant approval was obtained from the University Ethical Review Committee. An introduction letter was obtained from the Department of Biology, FUTA to seek the consent of participants before blood samples were collected.

Design

The sample population consisted of one hundred and eighteen (118) females and one hundred and eleven (111) males respectively. A total of two-hundred and fifty (250) individuals volunteered for the procedure. However, a total of two-hundred and twenty-nine (229) respondents correctly completed the questionnaire and were registered before sampling began and considered for the experiment after relevant demographic data were obtained using a well-structured questionnaire which covered gender, age, attitude towards use of Insecticide treated

nets (ITNs), previous exposure to malaria and syphilis etc. The study was conducted from March to September, 2021.

Sample Collection

Two millimetres (2 ml) of blood was collected from the subjects by venipuncture using sterile needle and syringes. Blood collected were dispensed into ethylene-diamine-tetra-acetic acid (EDTA) anticoagulated blood containers, properly mixed and labelled appropriately.

Determination of ABO and Rhesus factor

The ABO blood group of each subject was determined using cell grouping Antisera (A, B, AB and D). Four (4) drops of each subject blood sample is placed on separate points on a sterile white tile divided into four (4) cells. A drop of antisera A, B, AB and D were placed beside the blood and thoroughly mixed to obtain a homogenous mixture with the aid of a sterile rod and the tile was rocked gently to ensure uniform mixing. The mixtures were carefully observed to determine blood group of students by the presence of agglutination or not. Antiserum D was used to determine the Rhesus factor. Finally, the blood samples of respondents were grouped based on these observation and confirmation into blood group A+, A-, B+, B-, AB+, AB-, O+ and O- (Simon-oke *et al.*, 2016).

Screening for Treponema Antigen

Rapid test for syphilis was done to qualitatively detect the presence of antibody IgG and IgM to *Treponema pallidum* antigen in the whole blood of sampled population. Using whole blood sample, a drop of blood was placed on the test kit and two drops of buffer were added and observed within 15 minutes to detect a positive or negative test reaction.

Parasitological Examination and Quantification of *Plasmodium sp.* in blood samples

Malaria parasites were identified by examining the blood samples under a microscope. The presence or absence of Plasmodium parasite was determined by the preparation of thick and thin smears from the samples collected.

Thick and thin films were prepared on separate clean and grease free slides after appropriate labelling and allowed to air dry. Thin films were done with a spreader at an angle of 45° and fixed with methanol to evaporate while a smear of about 1-2 cm size of a dime was made for thick films which was allowed to dry and not fixed with methanol. Slides were arranged on a staining rack and flooded with 10% Giemsa Stain solution for 10 minutes, washed under a slow flowing tap water and allowed to air dry. The purpose of staining is to give the parasite a distinctive appearance. Finally, the films were carefully examined under oil immersion microscope objective (x100) and parasitaemia was calculated per 200 White blood cells.

Data Analysis

Data obtained were subjected to Chi-square test and ANOVA (SPSS version 20) to assess the difference between frequencies (the relationships between blood groups, *Plasmodium species* and Syphilis) and analysed at 95% level of significance. Observed difference was considered to be significant for $p < 0.05$.

$$\text{Prevalence} = \frac{\text{Total number of infected individuals}}{\text{Total no of examined individuals}} \times 100\%$$

3. RESULTS

Result revealed that 172 (75.1%) were positive for malaria parasite, distributed across blood group O 69(40.1%), A 39(22.7%), AB 34(19.8%) individuals; while the least infected was group B with only 30(17.4%). The relationship between ABO blood group and malaria parasite prevalence was significant in the study population ($P = 0.002$, $p < 0.05$); however, Statistical analysis showed no significant association between malaria parasite, ABO Blood groups and syphilis ($P > 0.05$, $P = 0.15$).

According to gender prevalence for malaria infection, there was no significant relationship as parasite prevalence was equally distributed between male and female subjects respectively ($X^2 = 1.767$, $P > 0.05$, $P = 0.184$). There was a slight difference of 1% in the male gender as compared to females (see Fig. 1).

From this finding, the prevalence of malaria parasite infection among age-group was not statistically significant; however, 55% of subjects between ages 20-24 were mostly predisposed among others (see Fig. 3) where the males (51%) recorded higher percentage compare to the female students ($X^2 = 5.62$, $P > 0.05$, $P = 0.06$).

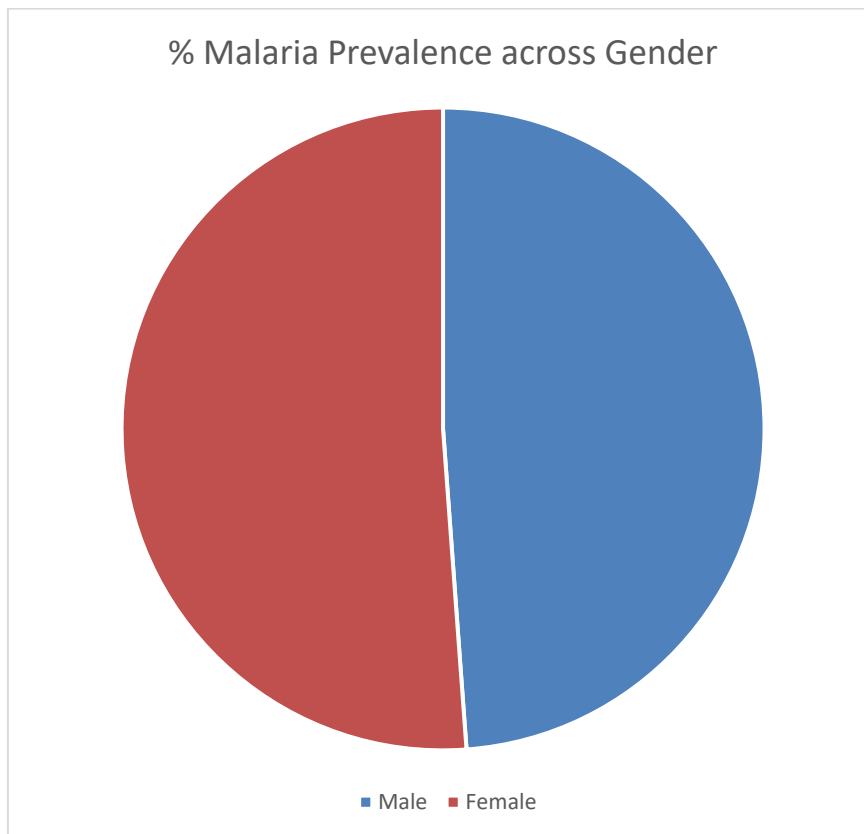


Figure 1. Malaria Prevalence across gender

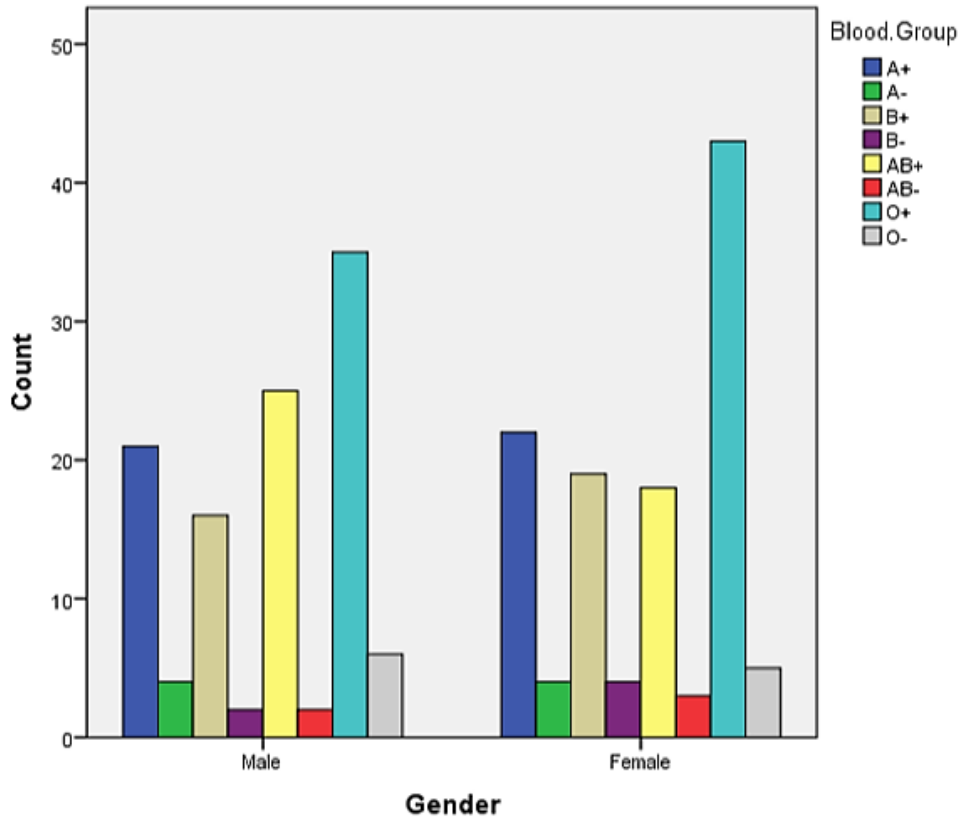


Figure 2. Blood group prevalence across gender

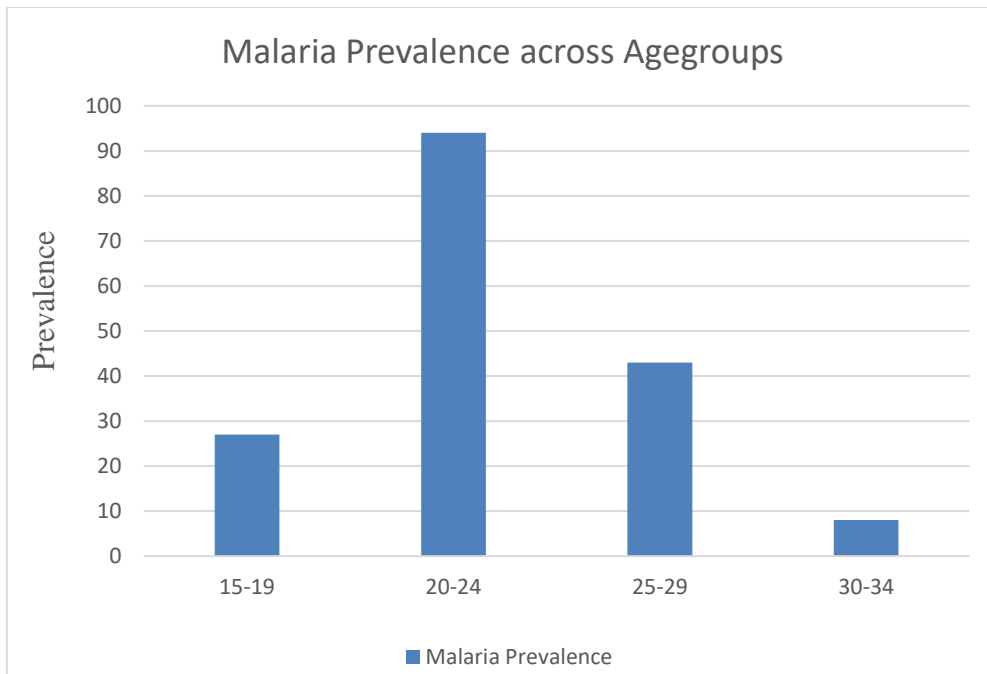


Figure 3. Malaria Prevalence across Age-groups

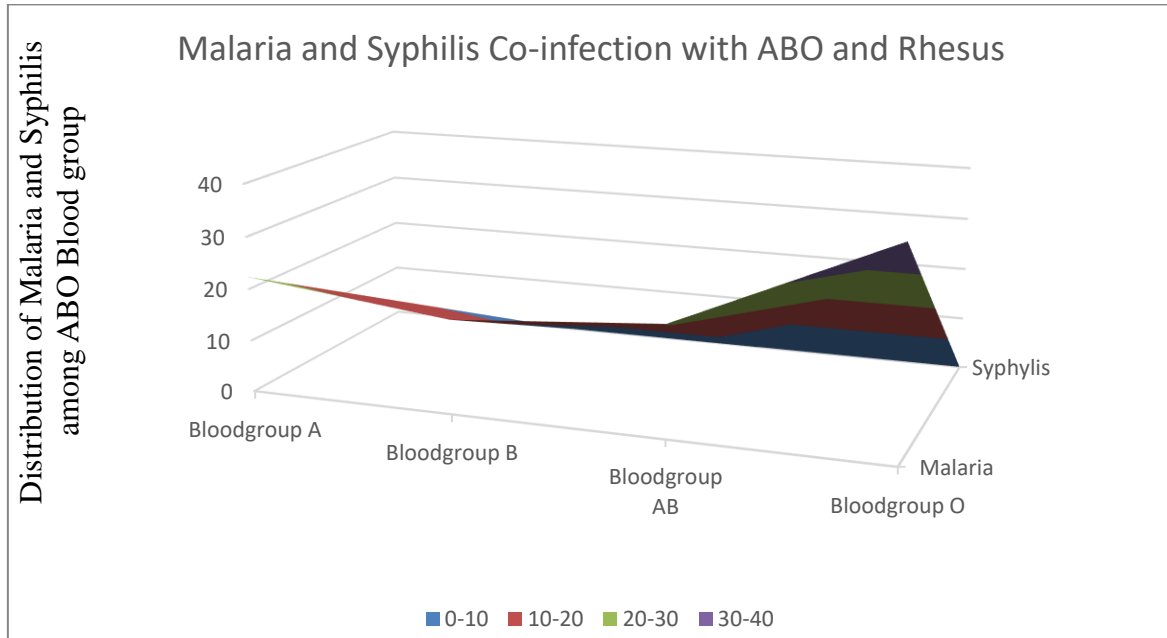


Figure 4. Distribution and Co-infection of Malaria and Syphilis with ABO and Rhesus

4. DISCUSSION

The study gathered that the blood type O (89) dominated the chart out of 229 subjects while blood group B (41) recorded the least prevalence among the sampled population for ABO grouping (see Fig 2); where 87% were rhesus positive and only 17% were discovered to be Rhesus negative in the study population. This may be due to tribal relations in the frequency of ABO blood group genes. The distribution of ABO blood group may be impacted upon by the prevalent tribe in an area or geographical location and so exhibit racial patterns in distribution.

The reports of Faduyile *et al.* (2016) stated blood group AB as the least occurring, which is in contrast as compared to this finding. More so, Rhesus factor negative may be an important factor to be considered even when minimal, as ignorance of this may result into medical complications especially congenitally related (Otajevwo, 2013).

Malaria parasite prevalence was least recorded among blood group B. This is not similar to the findings of Sule *et al.*, 2014 that showed higher prevalence of malaria among its study population. However, plasmodium infection seemed to be relatively high across blood group O (40.1%) and these differences may be due to the varying ethnic, racial and geographical background of the studied population; which is further buttressed by the reports of Goel *et al.*, 2015, that because of endemicity of malaria in Nigeria, more than half of the population belongs to blood group O.

This finding further revealed that out 229 students sampled, 172 (75.1%) harboured plasmodium parasites of varying intensities in their blood stream, and this proposes holo-endemicity of malaria among the students of FUTA as stated in the World Health Organization (WHO) standard for classification of endemicity.

This research highlighted that the prevalence of malaria is high in the study population and is an indication that the students are exposed to mosquito bites which may be due to

deficiency in the use of Insecticide Treated Nets (ITNs) and other unconscious, carefree attitude towards malaria preventive and control measures (Simon-oke *et al.*, 2016).

Gender specific prevalence showed that male and female students were equally at risk of getting infected with malaria parasite but not syphilis. This could imply that students heed to necessary sexual practices hence the reason for zero prevalence cases of *Treponema pallidum* in the study population. Although, malaria prevalence was slightly higher in male (75.67%) than female (74.5%). However, higher prevalence based on sex at particular age have been previously documented by Austin *et al.*, (2014) stating that prevalence of malaria based on gender is compounding to determine. Furthermore, higher malaria prevalence recorded in males could also be due to nature of males going 'shirtless' and exposing their bodies during hot weather which may subsequently increase chances of being exposed to mosquito bites (Akanbi *et al.*, 2010).

Garba *et al.* (2016) reported that variation in prevalence rate of malaria based on location could be associated to differences in *Anopheles* species, environmental and climatic conditions, study period, lifestyle of study population and diagnostic test methodology. Oladeinde *et al.* (2014) reported higher prevalence of malaria in male, while Okonkwo *et al.* (2010), Tela *et al.* (2015) reported higher prevalence of malaria in females which is in contrast to this present study. In addition, malaria prevalence could also be attributed to rainfall, warm temperatures, and stagnant waters which provide ideal habitats for mosquito larvae (Ukaegbu *et al.* 2014).

This study also suggested that malaria parasite could be age-related however, there was no significant statistical relationship among malaria parasite infection and age-group. Meanwhile, it is obvious from this study that the majority of the infected individuals were the young adult (20-24), which is in line with World Health Organization (WHO) age-group classification.

The sample population were physically healthy student, where 75.1% were positive for malaria parasite infection. The holo-endemicity of afebrile malaria among the university students of FUTA may result in severe morbidity with varying intensity. This may cause serious damage if not adequately attended to as it has potential for a greater risk of higher transmission in future (Prusty *et al.*, 2021). For the purpose of improving malaria burden and anaemia which impairs cognitive development in Africans, there should be targeted intervention for prompt diagnosis, treatment and control as part of school health policy (Makenga *et al.*, 2022).

5. CONCLUSIONS

Malaria is holo-endemic (75%) in the study population. This calls for promptness in proper diagnosis and treatment of malaria among students, to prevent upsurge in emergency cases. Most especially, the asymptomatic malaria (ASM) whose potential of causing critical febrility crisis and morbidity should be checked in order to curb malaria casualties among university students. The zero prevalence of syphilis in the sampled population showed that they were conscious of their sexual health and took adequate measures when selecting a sexual partner. However, emphasis must be made on adequate testing for sexual partners in the society, as people tend to shun and sometimes disallow knowing about their sexual health status. The high prevalence of malaria parasite encountered, further established that socioeconomic factors, such as appalling unsanitary condition, low standard of living, inadequate use of treated mosquito nets are still very much in play as a predisposing factor to malaria infection in the

study area. Lastly, the non-statistically significant relationship among ABO, Malaria and syphilis in study population may suggest that both males and females are equally at risk under any given circumstances.

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