



Prevalence of Transfusion-Transmissible T. Pallidum Infection and Coinfection with HIV, HBV and HCV among Voluntary Blood Drive Donors from Selected Senior High Schools in Tamale

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Abstract

Syphilis is a sexually transmitted disease caused by the bacterium Treponema pallidum and is a current systemic disease of public health concern. In 2022, global estimates of syphilis cases reached 700,000 with widespread consequences including 150,000 perinatal deaths and 70,000 neonatal deaths. Compared to global research, little has been done to assess and synthesize the disease trends within high-risk categories in sub-Saharan Africa including Ghana. This cross sectional survey evaluated the seroprevalence of syphilis and its coinfections with HIV, HBV, and HCV among blood drive donors from selected Senior High Schools (SHSs) within the Tamale metropolis who form a sexually active risk group. The study included 119 donors between 17 and 25 years who consented and met eligibility criteria of the National Blood Service, Ghana. 5 ml of each donor's blood sample was aliquoted into an EDTA-coated test tube and centrifuged after which the serum obtained was screened for syphilis, HIV, HBV and HCV using the respective Fortress Diagnostics ELISA test kit. Data obtained was analysed using SPSS version 26. Figures were constructed with GraphPad Prism 10. A seroprevalence of 4.2 % was obtained for syphilis among subjects. Of all syphilis positive cases, 20 % had a coinfection with HIV suggesting that syphilis may facilitate the transmission of HIV. Syphilis was the most predominant infection among subjects compared to HIV, HBV, and HCV. There was a significant gender disparity in the trend of infection with more males testing positive for syphilis than females, suggesting that males are at a higher risk of contracting STIs. Our data underscores the need to target second cycle institutions for public health interventions as these infections are circulating among youngsters in SHSs and are at high risk of syphilis infection.

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List of abbreviations

ASIR:	Age-standardized incidence rate
HBV:	Hepatitis B virus
HCV:	Hepatitis C virus
HIV:	Human Immunodeficiency Virus
HRP:	Horseradish peroxidase
MSM:	Men who have sex with men
SHSs:	Senior High Schools
TTH:	Tamale Teaching Hospital
WHO:	World Health Organization
VNRDs:	Voluntary non-remunerated donors

Introduction

Syphilis is an ulcerative sexually transmitted infection (STI) which is caused by the spirochaete *Treponema pallidum* (*T. pallidum*), subspecies, *pallidum*, and belongs to the genus *Treponema* [1]. It is a systemic disease of major global public health concern (more so when coinfecting with human immunodeficiency virus, HIV) because of its high prevalence, infectiousness, and toll on both infected individuals and health systems across the world [2]. The infection is a multi-stage condition (comprising primary, secondary, latent and tertiary syphilis) that progresses over many years if left untreated. Each of the 4 phases of syphilis is characterized by differential signs and symptoms and is implicated in increasing risk of coinfection with other STIs such as HIV, hepatitis B virus (HBV), and hepatitis C virus (HCV) among others [3,4].

The disease may be transmitted horizontally by contact with infected skin lesions (chancre, mucus patch, or condyloma latum) during oral, anal or vaginal sex. The disease may also be acquired vertically, from mother to fetus mainly through transplacental transmission during pregnancy (i.e., congenital or prenatal transmission) or, less commonly, during

delivery through contact with a maternal lesion (con-natal transmission) [4,5]. Additionally, newborns may contract the disease during breastfeeding via contact with maternal nipple lesions. The disease may also be transmitted through blood transfusion as well as par-enteral transmission among drug addicts [6-9].

Previously, arsphenamine, mercury, purgatives, heat and pyrogens have been used to manage the disease [10]. Penicillin has been successful for the treatment of syphilis as *T. pallidum* has not developed resistance to penicillin since 1943 and is still highly recommended. However, 23S ribosomal RNA gene mutations, A2058G that confer resistance to macrolide antibiotics (across North America, Europe and Asia) and A2059G (across United States, China, Australia, the United Kingdom and the Czech Republic) have been identified [11]. The standard course of treatment for syphilis in all stages is still benzathine penicillin G [12,13]. More recently, medical interventions involving the use of doxycycline as a pre- and/ or post-exposure prophylaxis have demonstrated substantial efficacy and tolerability against syphilis among other STIs even though its use is controversial at this stage [14,15]. However, despite availability of treatment, the disease prevalence continues to surge in resource-limited settings [2] and recently in the developed world where it was previously thought to have been eradicated [1,6,17].

The most important risk factors that influence syphilis transmission include unprotected sexual activity that involves contact with oral, genital or anal mucosa, particularly anal sex among men who have sex with men (MSM) [18,19]. Having multiple partners, or sex with a partner who has tested positive for chlamydia, gonorrhoea, herpes simplex virus (HSV-2) or who has a past history of syphilis infection increases risk of contracting the disease [18]. Substance use,

including chemsex (the use of drugs e.g., methamphetamine, mephedrone, gamma-hydroxybutyrate, gamma butyrolactone), using the internet to access sexual contacts, and group sex have also been reported as risk factors [18,20]. Additionally, a strong correlation between HIV infection and transmission with syphilis has been demonstrated [16,18,21].

Syphilis has a high morbidity rate, making it a prominent subject of concern. If treatment is not administered, the tertiary phases of syphilis may lead to neurological, cardiovascular, and dermatological disease in adults [22,23]. If the infection spreads during the first trimester of pregnancy, potential deleterious outcome may be stillbirth, low birthweight (<2.27 kg), nonimmune hydrops, fetal loss (miscarriage), premature delivery, hearing loss, blindness, neonatal death or severe congenital abnormalities in infants [4,24]. Infected individuals often suffer stigmatization, stereotyping, vulnerability, shame, and gender-based violence [21]. Syphilis may also in turn raise the risk of developing other STIs, such as HIV [25,26].

Global burden estimate of syphilis cases in 1990 was 30.91 million and 49.71 million in 2019, a 60.83 % rise within the period [27]. The age-standardized incidence rate (ASIR) increased from 8,845,220 (95 % UI: 6,562,510 – 11,588,860) in 1990 to 14,114,110 (95 % UI: 10,648,490 – 18,415,970) in 2019 and 160.03/100,000 persons (95 % UI: 120.66 – 208.1) to 178.48/100,000 persons (95 % UI: 134.94–232.34), respectively with an estimated annual percentage change in ASIR of 0.16 (95 % CI: 0.07–0.26) [28]. The World Health Organization (WHO) estimates for 2016 indicate a global prevalence of 0.5 % in syphilis cases (95 % CI: 0.4 % – 0.6 %) in both men and women of which most congenital cases (61 %, ~ 404,000) occur in African [29,30]. Treponemal infection prevalence in Africa are varied [2,31,32]. In south Africa, Hoque et al. estimated prevalence of syphilis to be 3.8 %. Farahani et al. estimated a prevalence of 0.9 % in Tanzania and Zimbabwe, 2.1 % in Uganda, and 3.0 % in Zambia [33,34]. Syphilis prevalence was higher among people living with HIV ranging from 2.6 % in Ethiopia to 9.6 % in Zambia compared with HIV negative individuals. In Ghana prevalence of syphilis in various subpopulations and in different regions is pegged

between 0.3 and 19.1 % [35-37].

T. pallidum is one of the earliest infectious risks associated with blood transfusions which ultimately led to the development of WHO standards necessitating *T. pallidum* screening in blood donors [38]. Global database on blood safety indicate about 1.6 million units of blood out of 92 million donated worldwide is discarded due to infectious signs of treponematoses [39,40]. These recommendations have drawn criticisms since a large number of syphilis antibodies among blood donors may result from antibodies acquired from past infections or from non-specific reactions. Moreover, *T. pallidum* is heat labile (thriving at 20 – 40 °C) and cannot tolerate cold storage temperatures of 2 – 6 °C either [41]. Seroconversion and syphilis seropositivity are highly prevalent in blood donors and so recipients of blood transfusions are at risk of contracting transfusion-transmitted syphilis where screening is not performed [6,42, 43]. Reported treponemal infection prevalence among blood donors in Africa is between 0.71 to 20 % [44]. In Ghana, prevalence among seemingly healthy blood donors and pregnant women are 3.7 % and 0.3 % respectively [45]. This result is probably found at blood banks where there is a high demand for blood resulting in short blood storage periods. Indeed, investigations indicate survival time of *T. pallidum* in banked blood is ≥ 72 hr, consistent with that reported by WHO [46]. Adegoke and Akanni called for a major review of the practice of screening donor blood in Nigeria earlier, stating that survival period of *T. Pallidum* in banked blood is 72 – 120 hr. Owusu et al. also demonstrated the need for donor blood syphilis screening and highlighted transfusion-transmitted syphilis as a serious public health problem in developing countries [47-48]. Additionally, as investigations provide evidence of strong coinfection of syphilis with other STIs, collateral testing for HIV-1 and -2, HBV, HCV and other STIs on patients diagnosed with syphilis is highly recommended [49,50].

The highest risk population includes youngsters aged 20 – 35 years and young mothers between 35 and 39 years [51,52]. However, there has been an increase in cases recently, particularly in people <25 years [53]. Every year, approximately 6 million new cases of syphilis infection is reported globally in persons aged 15 to 49 years. Additionally, >300,000 fetal and neonatal deaths with >215,000 infants placed at increased

risk of early death are attributable to syphilis infection [2,50]. The WHO reports indicate syphilis affected an estimated 8 million persons worldwide between 15 and 49 years in 2022 and ~1.1 million pregnant women were diagnosed with syphilis which resulted in over 390,000 adverse birth outcomes [54]. Such estimates provide evidence and reference data for targeted interventions, prevention, care programme improvements, monitoring and evaluation [55-57] including drug development (e.g., vaccines) [58,59]. However, there is still limited literature to describe the true burden of syphilis, the prevalence, incidence, seroconversion, associated risk factors, *T. pallidum* strain types and occurrence of new variants in Ghana. Although there is a vast quantity of research on the epidemiology of syphilis conducted globally, not much has been done to synthesize the body of knowledge and rigorously assess disease trends within high-risk categories in Ghana, much less meta-analysis. We therefore aimed to determine the seroprevalence of *T. pallidum* infection among young adult blood donors (who form part of the high-risk group) from selected SHSs within the Tamale metropolis and identify subgroups at higher risk of syphilis infection. The study was also designed to assess coinfection with HIV, HBV and HCV among the study population. The study was conducted in collaboration with the blood drives organized by the blood bank of the Tamale Teaching Hospital across schools within the metropolis.

Materials and Methods

The study was conducted at the TTH, Tamale, which is the northern regional capital of Ghana. The study was carried out in collaboration with the blood bank during blood drive sessions between January and July 2024. The study population included 119 students between 17 and 25 years from selected SHSs within the metropolis who met the inclusion criteria for blood donation according to the guidelines of the National Blood Service, Ghana, and consented to donate blood during blood donation drive session by filling a questionnaire/donor recruitment form from the service. The Cochran formula ($N = Z^2 PQ / D^2$) was used to ascertain the sample size, where *N* represents the desired sample size; *Z* (1.96), the standard normal deviation at a confidence interval of 95 %, *D* (0.05), the degree of accuracy and *P* (8.5 %), the estimated prevalence of syphilis in

Ghana [60], whereas $Q = (1 - P)$.

450 mL of blood was drawn from each participant through venipuncture by qualified donor nurses and aliquoted into EDTA test tubes to be used as test samples. These samples were centrifuged at 4,000 rpm for 5 min after which supernatant containing plasma was serologically screened for presence of anti-*T. Pallidum* (anti-TP), anti-HIV, anti-HBV and anti-HCV.

Detection of Anti-TP in Blood Samples

A 96-Well Fortress Diagnostics ELISA (Enzyme-linked immunosorbent assay) syphilis test kit (Fortress Diagnostics Ltd, Antrim, UK) with a sensitivity >95 % and specificity >99 % was used to detect anti-TP following manufacturer's instructions. Reagents were allowed to reach RTP (18 – 30 °C) for 15 – 30 min. 3 negative controls, 2 positive controls and 1 blank were added to the experiments. First, a 100 µL of Horseradish peroxidase (HRP) conjugate was added to each test well except the blank, to which 100 µL of distilled water was added. To each test well, 20 µL of appropriate plasma sample was added while an equivalent volume of positive and negative controls was added to their respective wells. The reaction mixtures were incubated at 37 °C for 60 min. Following this incubation period, each well was washed 6 times with phosphate buffer to remove unbound proteins and blotted dry using a Whatman filter paper. 50 µL of chromogen A and B solutions were then added to each well, mixed and incubated at 37 °C. This step is critical for the determination of any change in colour. After 15 min of incubation, the reactions were stopped by addition of 50 µL of acid-based stop solution to each well after which the absorbance was read at 450 nm and the concentrations determined using the Chemwell-Fusion-Automated Enzyme Immunoassay (EIA) and Chemiluminescence Immunoassay (CLIA) analyzer (Awareness Technology, Inc., Palm City, USA). Results were then interpreted as negative if concentration <1 and positive if concentration ≥1.

Detection of HIV in the Blood Samples

The ELISA test kit for HIV from Fortress Diagnostics (Fortress Diagnostics Ltd, Antrim, UK) was used to detect anti-HIV antibodies following the manufacturer's instructions. Briefly, reactions were set up as described for syphilis. After equilibrating samples and reagents at RTP for 15 – 30 min, 20 µL of biotinylated

anti-HIV p24 antibodies was added to every well except the blank which contained 20 μ L of double distilled water. 100 μ L of positive control, negative control, and plasma samples were then pipetted into their respective wells, mixed and incubated at 37 °C for 60 min. After incubation, each well was washed 6 times with a wash buffer and blotted on Whatman filter paper. 100 μ L of HRP conjugate was then added to each well and further incubated at 37 °C for 30 min after which wells were washed and blotted as previously described. 50 μ L of chromogen A and B solutions were then added to each well including the blank, mixed, and incubated at 37 °C for 15 min for colour to develop. The reactions were then stopped by adding 50 μ L of stop solution to each well and mixed. Absorbance was read at 450 nm, concentrations determined, and results interpreted as, negative if concentration is <1 and positive if concentration is $\geq$ 1 as previously described for syphilis.

Detection of HBV in Blood Samples

The ELISA test kit for HBV from Fortress Diagnostics (Fortress Diagnostics Ltd, Antrim, UK) was used to detect HBV. Reactions were set up as previously described for syphilis. After equilibrating samples and reagents, a 100 μ L of HRP conjugate was pipetted into each well except blank in which 100 μ L of double distilled water was pipetted. 50 μ L of positive control, negative control, and plasma were then added to their respective wells and incubated at 37 °C. After 60 min of incubation, reaction wells were washed 6 times and blotted on paper as described previously. Colour was developed by adding 50 μ L of chromogen A and B solutions to each well including blank, mixed, and incubated at 37 °C for 15 min after which reactions were stopped, absorbance measured, and results interpreted as described previously for syphilis.

Detection of HCV in Blood Samples

Reactions were set up as for HBV. 100 μ L of specimen diluent was added to each labelled well after which 10 μ L of positive control, negative control, and plasma samples were added to appropriate wells. Reaction mixtures were mixed and incubated at 37 °C for 30 min. Each well was then washed and blotted on filter paper as previously described after which 100 μ L of HRP conjugate was pipetted into each well and re-incubated at 37 °C for 30 min.

Colour was developed and reactions were stopped as described previously. Absorbance was then measured, and results interpreted as previously described.

Determination of Blood Groups of Participants

The test tube method was employed to determine blood groups of the participants using monoclonal grouping sera; Anti-A, Anti-B and Anti-D/Rhesus (ARKRAY Healthcare Pvt. Ltd., Mumbai, India). Anticoagulated blood from each blood sample aliquoted for testing was washed with normal saline after which a 5 % cell suspension of each was made. A drop of grouping sera was added into their respectively labelled tubes, Anti-A, Anti-B, Anti-D and Auto. 2 drops of participant's plasma were added to the tube labelled Auto after which equal volumes of the 5 % cell suspension were added to each tube and mixed. The anti-sera cell suspension mixture was centrifuged at 1,000 rpm for 1 min and examined for agglutination by gently shaking the tube to resuspend the cells. The results were interpreted as follows: No agglutination in any tube was reported as blood group O, agglutination in Anti-A tube only was reported as group A, agglutination in Anti-B tube only was reported as group B, agglutination in both Anti-A and Anti-B tubes was reported as group AB. The Rhesus typing was interpreted as follows: an agglutination in the Anti-D tube indicated Rh-positive and no agglutination indicated Rh-negative.

Data Analysis

The Statistical Package for Social Sciences (SPSS) (IBM, NY, USA) version 26 was used to analyze the data. GraphPad Prism 10 (GraphPad Company, Boston, USA) was used to construct graphs. Data generated was presented with descriptive statistics.

Results

Demographic Characteristics

A total of 119 students who voluntarily donated blood to the mobile blood bank unit of the TTH from 3 second cycle institutions were recruited for study. Blood was obtained from each donor after which each blood sample was tested for syphilis, HIV, HBV, and HCV using an enzyme-linked immunosorbent assay. Majority of participants (40, 33.6 %) were 18 years (Figure 1). The subgroup with the least number of participants (1, 0.8 %) was 23 years. More males (77.31 %) than females (22.69 %) participated in the investigation

(Figure 2). Most donors (42.0 %) were of the O positive blood type (Table 1). The least of the participants (1.7 %) were of the A negative type. O negative universal donor blood type identified among participants were also few (3.4 %).

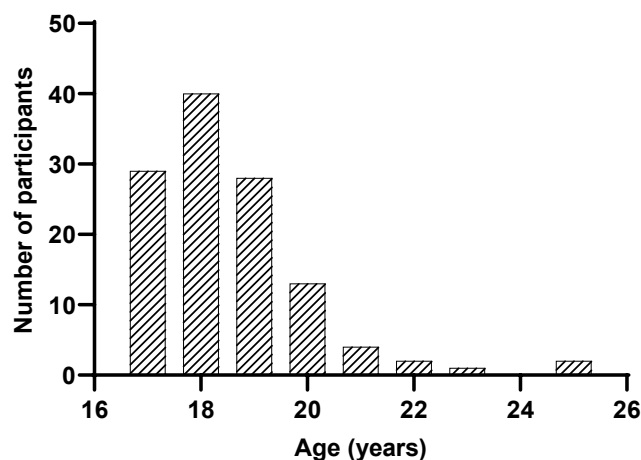


Figure 1: Age distribution of Participants.

Of the 119 student participants between 17 and 25 years, 29 were 17 years representing 24.4 %, 40 (33.6 %) were 18 years representing the subgroup with highest number of participants, 28 (23.5 %) were 19 years, 13 (10.9 %) were 20 years while 4 (3.4 %) were 21 years. Respectively, 2 (1.7 %), 2 (1.7 %) and only 1 (0.8 %), the subgroup with the lowest number of participants were 22, 25 and 23 years. No participant was 24 years. The mean age of recruits was 18.54 years with a standard deviation of 1.5 years. Expectedly, most participants were between 17 and 19 years as correct age of students in SHS is 17 – 19 years before completion.

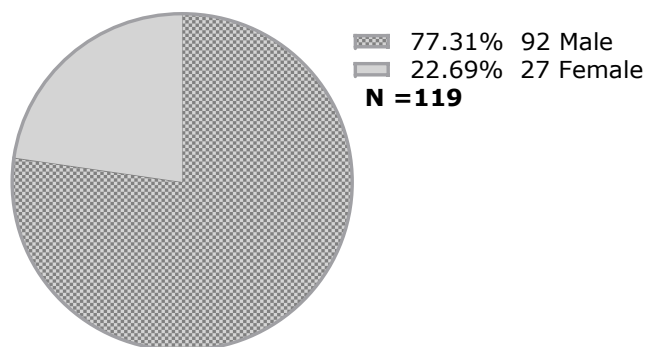


Figure 2: Sex Distribution of the Participants.

Most participants were male representing 77.31 % (92). Females were only 22.69 % (27) of total.

Table 1: Donor Blood Groups

No	Blood group of participants	Number of participants	Percentage of total (%)
1	A negative	2	1.7
2	A positive	22	18.5
3	AB positive	3	2.5
4	B negative	3	2.5
5	B positive	35	29.4
6	O negative	4	3.4
7	O positive	50	42
Total		119	100

Serological Status of Participants

The computed seroprevalence of syphilis among blood drive donors from the selected SHSs who fall under the category of voluntary non-remunerated donors (VNRDs) is 4.2 %. The prevalence of syphilis per 1,000 of the study population is therefore 42 per 1,000 SHS students. The distribution of test results for syphilis, HIV, HBV, and HCV among the study population were varied. Of the number of recruits, 2 individuals (1.7 %) were positive for HIV, 3 (2.5 %) for HBV and only 1 (0.8 %) for HCV (Table 2). Of the 92 male participants, 4 (4.3 %) were positive for syphilis, 2 (1.7 %) for HIV, 3 (3.3 %) for HBV and none for HCV. Of the 27 female participants, 1 (3.7 %) was positive for HCV. No female examined had HIV nor HBV (Table 2, percentages of total are included in parenthesis).

Table 2: Laboratory Blood Analysis results of Participants

Age (years)	Sex	Syphilis		HIV		HBV		HCV	
		Positive	Negative	Positive	Negative	Positive	Negative	Positive	Negative
17	M	0	23 (19.3%)	1 (0.8%)	22 (18.5%)	1 (0.8%)	22 (18.5%)	0	23 (19.3%)
	F	0	6 (5.0%)	0	6 (5.0%)	0	6 (5.0%)	0	6 (5.0%)
18	M	4 (3.4%)	29 (24.4%)	1 (0.8%)	32 (26.9%)	2 (1.7%)	31 (26.1%)	0	33 (27.7%)
19	M	0	20 (16.8%)	0	20 (16.8%)	0	20 (16.8%)	0	20 (16.8%)
	F	0	8 (6.7%)	0	8 (6.7%)	0	8 (6.7%)	1 (0.8%)	7 (5.9%)
20	M	0	9 (7.6%)	0	9 (7.6%)	0	9 (7.6%)	0	9 (7.6%)
	F	0	4 (3.4%)	0	4 (3.4%)	0	4 (3.4%)	0	4 (3.4%)
21	M	0	3 (2.5)	0	3 (2.5%)	0	3 (2.5%)	0	3 (2.5%)
	F	0	1 (0.8%)	0	1 (0.8%)	0	1 (0.8%)	0	1 (0.8%)
22	M	0	2 (1.7%)	0	2 (1.7%)	0	2 (1.7%)	0	2 (1.7%)
	F	0	0	0		0	0	0	0
23	M	0	0	0		0	0	0	0
	F	0	1 (0.8%)	0	1 (0.8%)	0	1 (0.8%)	0	1 (0.8%)
25	M	0	2 (1.7%)		2 (1.7%)	0	2 (1.7%)	0	2 (1.7%)
	F	0	0	0		0	0	0	0
Total		5 (4.2%)	114 (95.8)	2 (1.7%)	117(98.3%)	3 (2.3%)	116 (97.5)	1 (0.8%)	118 (99.2%)

Discussion

Of the 119 students who participated in this study, the majority were 18 years old (40) representing 33.6 %. There was a gradual decline in donors with an increase in age with a mean age of 18.54 years. The median age of 18 years is consistent with several studies. The age range of donors in a study conducted in the United States by Kane et al. was 16 to 99 years, with 18 years as the most common age at donation [61]. Expectedly, most participants were between 17 and 19 years as the typical age of students in SHS is 16 – 18 years [62]. Students above this age range might have started school later than usual, stayed out of school for some time or might have returned to school after the Ghana Government introduced the free SHS policy which allows poor and needy students, and SHS leavers to return to school to sit or re-sit the West African Senior Secondary School Certificate Examinations (WASSCE) [63]. More males (77.31 %) than females (22.69 %) participated in the investigation (Figure 2), possibly due to the existing gender disparity in SHSs especially in the northern part of Ghana. It is also a reflection of the situation in the metropolis that more males than females reach SHS level, however, this situation is not peculiar to the Northern Region. It is a similar trend in most other parts of the country [64, 65]. Most donors (42.0 %) were of the O positive blood type which is critical in trauma care because they can only receive transfusions from O positive or O negative blood types (Table 1) in consonance with findings that O positive is globally the commonest blood group.

The High prevalence of syphilis (4.2 %) obtained in this study has been observed in several similar investigations across the world. For instance, a 3.1 % seroprevalence of syphilis among manual workers has been reported by Alharazi et al. [39] in Yemen. In Ethiopia, the seroprevalence of syphilis among sex workers investigated was reported to be 6.2 %, of which the 20 – 24-year-old subgroup was the majority in line with the current study [66]. This result is only slightly higher possibly because sex workers constitute a very high-risk group. Meta-analysis of 24 different syphilis seroprevalence studies among Chinese High School and College male homosexual student population is also relatively higher (5.7 %) possibly because the study strategically targeted

MSM who also form part of the highest risk group [67]. The seroprevalence of syphilis in the High Schools and Colleges in Chinese major cities investigated ranged from 2.2 in Liaocheng to 7.8 % in Beijing [67].

In Ghana, comparatively high seroprevalence of 3.2 % was obtained among outpatients 15 – 49 years in the Asikuma Odoben Brakwa District with more positive males (5.7 %) than females (1.7 %) [68] in consonance with the current study. The slight difference might be due to the higher sample population used in this study with respect to our study [68]. More recent retrospective studies of transfusion-transmissible infections from across other regions in Ghana include remarkably high syphilis seroprevalence of 8.9 % in Offinso-North District, Ashanti Region, and with a 19.1 % coinfection with HIV [49], 15.3 % in Koforidua, Eastern Region, with 0.2 % coinfection with HIV and 6.8 % from combined secondary data obtained from 4 regions; the Greater Accra, Oti, Northern, and Upper West [69,37]. However, our result may be higher than 1.3 % previously obtained by Tessema et al. in a similar study among blood donors done on a much larger study population in Ethiopia [70]. This may be because the current study focused primarily on students between 17 and 25 years who are a high-risk age category, are vulnerable and more likely to engage in high-risk sexual behavior due to lack of awareness, naivety and limited access to healthcare facilities. According to Musonda et al. overall mean age at first sex among adolescent girls and young women in Zambia interviewed is 16.6 years and so are vulnerable at 17 – 19 years. Similarly, the prevalence of syphilis was high of the order of 4.1 % among girls out of school compared with girls in school [71]. As such, we may speculate that prevalence of youngsters between 17 and 25 years who are out of school in Tamale, might be much higher than 4.2 % found in the current study.

Our Results suggest that whereas HCV exhibited a minor incidence among females, HIV, HBV, and syphilis were more common in males than in females (Table 2). That is, more male students (4, 3.4 %) than females (1, 0.8 %) were positive for syphilis possibly because more males (92, 77.3 %) than females (27, 22.7 %) participated in the investigation and therefore the probability of having a positive case who is a male was much higher than that of females. However,

this may also indicate that male students are at a greater risk of contracting syphilis and potentially other STIs compared to their female counterparts and may extrapolate to the entire Tamale metropolis. This gender disparity may also be attributed to behavioural factors, such as higher engagement in risky sexual activities among males, or socio-cultural influences that affect access to health education and services.

The highest proportion of positive cases were 18 years. This may be because it constitutes the subgroup with the highest number of participants and/or indicative that this subgroup is more sexually active. Specifically, all 5 positive cases of syphilis and the 1 positive HIV case were 18-year-olds. Among the 17-year-subgroup, there was no positive incidence of syphilis nor HCV, however, 1 participant (0.8 %) tested positive for HBV and another (0.8 %) for HIV. Students ≥ 19 years were less likely to have any disease and again may be attributable to decreasing number of volunteers with increase in age (Figure 1). As such there was only 1 (0.8 %) positive case of HCV in the 19-year-old group whereas no positive cases of syphilis, HIV, HCV, or HBV was reported in the 20 – 25-year-old participants.

Our results show that the prevailing diseases among the selected infections in decreasing order are syphilis (4.2 %) > HBV (2.5 %) > HIV (1.7 %) > HCV (0.8 %). The few numbers result from a small sample size as the study was limited in many ways including funding and students' unwillingness to donate blood. This trend of infections suggests that syphilis is relatively more common among SHS students compared to HIV, HBV and HCV. It also implies that syphilis may be spreading among SHS students in Tamale and in the entire metropolis compared with the other infectious agents accessed here which necessitate targeted public health interventions. This notwithstanding, the fact that HBV and HIV have also been isolated from some samples indicates that these viruses are also circulating among youngsters in second cycle institutions and within the metropolis and requires more attention from policy makers. In addition, the study shows that more male than female voluntarily donate blood. This is comparable with several studies [55,56,72]. More importantly, the fewer VNRDs in the current study compared

with family donors (FDs) and replacement donors (RDs) who are often associated as family replacement donors (FRDs) are a bulk of blood donors in resource poor countries usually for cultural and economic reasons [72,73]. This may also explain the lower turnout than expected since there was no remuneration.

Of the 5 positive cases of syphilis, 1 (20 %) was coinfecting with HIV close to findings by Nkansah et al. in the Ashanti Region [41]. This is a critical finding as it suggests that syphilis may facilitate the transmission of HIV, a phenomenon that is well documented in medical literature. A strong correlation between HIV infection and transmission with syphilis has been reported by several researchers [16, 21]. This may be due to syphilitic vaginal ulcers of infected individuals which heavily infiltrate lymphocytes – essentially the primary target cells for HIV infection. As such lymphocytes may serve as the gateway for HIV acquisition and a hub for transmission of HIV (and syphilis) to others [74]. Individuals with syphilis are therefore at a significantly higher risk of acquiring HIV compared to those without the infection [74]. Moreover, studies have revealed that coinfection with syphilis can lead to higher concentrations of HIV in genital secretions, thereby enhancing the infectiousness of individuals with HIV [75]. Indeed, the tendency for coinfection with HIV and other STIs has made it possible to predict STIs (HIV, HBV, HCV, HSV-2) among *T. pallidum* infected individuals according to Ng'wamkai et al. [26].

Conclusion

Syphilis is apparently more prevalent among SHS students in the Tamale metropolis compared to other STIs (HIV, HBV, and HCV) in the current investigation, highlighting a need for targeted public health interventions. There is gender disparity in infection patterns with more males having STIs than females, suggesting that males are at a higher risk of contracting sexually transmitted infections, possibly due to behavioral factors such as higher engagement in risky sexual activities and socio-cultural influences that affect access to health education and services. The investigation also showed coinfections among individuals. A coinfection incidence of 20 % was observed for both syphilis and HIV. This finding is critical as it suggests that syphilis may facilitate the transmission of HIV. There is need to increase high-risk groups'

education (on STI prevention strategies such as condom use, behaviour counselling), particularly at second cycle institutions. Routine STI screening should be done in these institutions to enable early detection and management. It is also necessary to intensify campaigns on blood donations in the general public, to guarantee safety of blood through screening. Coinfections such as syphilis and HIV/AIDS among other STIs should be checked routinely.

Ethics Approval and Consent to Participate

The Department of Community Health and Preventive Medicine, School of Medicine, University for Development Studies, Tamale, and the Research Department of the Tamale Teaching Hospital, Tamale, both provided approval. For confidentiality, identifiable patient information was not captured in the study. Participants were identified by serial numbers.

Competing Interests

The authors declare no conflicts of interest.

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Authors' Contribution

SAN, CMS and JT

.D conceived the idea of the project. CMS and SAN did the laboratory analysis with inputs from JTD. SAN did the statistical analysis and JTD drafted the manuscript. All authors contributed to reviewing the manuscript.

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Data Availability statement

The integer data used to support the findings of this study are included within the article and openly available at <https://doi.org/>. [doi].

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