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Prevalence of Sexually Transmitted Infections (STIs) among Attendees of AFRH Centre in Ibadan, Southwestern Nigeria

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Abstract: In a cross-sectional study, 200 patients attending the Association for Reproductive Family and Health (AFRH) Centre in Ibadan, Nigeria from August 2011 to October 2011 were screened randomly to determine the prevalence of common sexually transmission infections (STIs) among them, using conventional methods. One hundred and ten (55.0%) of the subjects harboured various agents of STIs and 21 (10.5%) harboured other bacterial isolates. The study showed that *Candida albican* 54(27.0%) was the most predominant organisms among these subjects. This was followed by *Gardnerella vaginalis* 21(10.5%) and *Trichomonas vaginalis* 3(1.5%). Other isolates include *Staphylococcus aureus* 4(2.0%), *E. coli* 1(0.5%), coliforms 1(0.5%) and normal genital flora 15(7.5%). However, *Trepanema pallidium* (syphilis), *Neisseria gonorrhea* and *Chlamydia trachomatis* were not detected. Risk factors associated with significant STI were young age, sex and marital status. The importance of routine STI screening in sexually active patients especially among the young and singles is advocated. It is recommended that routine screening for STIs should be incorporated into hospital care.

Key words: *Candida albican* · *Gardnerella vaginalis* · *Trichomonas vaginalis* · STI · STD

INTRODUCTION

Sexually transmitted infections (STIs), including human immunodeficiency virus (HIV), continue to present major health, social and economic problems in the developing world, leading to considerable morbidity, mortality and stigma [1]. The prevalence rates apparently are far higher in developing countries where STI treatment is less accessible [1-2]. Many organisms such as *Neisseria gonorrhea*, *Chlamydia trachomatis*, *Gardnerella vaginalis*, *Treponema palladium*, *Trichomonas vaginalis* etc. has been incriminated as etiological agents of Sexually Transmitted Diseases (STDs) [3].

Sexually transmitted diseases (STDs) are a group of infectious or communicable diseases in which the primary mode of transmission is through sexual contact [4] and are among the major causes of illnesses in the world especially in the developing countries [5-7]. The diseases caused by STDs are classified according to the type of organism causing the infection, which could be bacterial, fungal, viral or of parasitic origin [8]. Some of the common sexually transmitted diseases include: *Bacterial vaginosis*, herpes, Chlamydia, trichomoniasis, gonorrhoea and syphilis [8]. More than 25 infectious organisms are transmitted primarily through sexual activity and studies reveal that STDs are among the many related factors that affect the broad continuum of reproductive health [9].

Sexually transmitted infections (STIs), continue to present major health, social and economic problems in the developing world, leading to considerable morbidity, mortality and stigma [1]. The prevalence rates apparently are far higher in developing countries where STI treatment is less accessible [1-2]. Most of the STIs, both ulcerative and nonulcerative, are prevalent in Nigeria and constitute one of the major public health problems. Their profile varies with changes in socioeconomic, cultural, geographic and environmental factors prevalent in

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different parts of the country [10-14]. However, due to lack of adequate laboratory infrastructure in the country, information regarding the profile of STIs relies essentially on syndromic diagnosis. Hence there is very limited data of laboratory-proven STIs [15-16]. However, the availability of baseline information on the epidemiology of STIs and other associated risk behaviors remains essential for the designing, implementing and monitoring successful targeted interventions [17-18].

The availability of baseline information on the epidemiology of sexually transmitted infections (STIs) and other associated risk behaviors is essential for designing, implementing and monitoring successful targeted interventions [1]. Also, continuous analysis of risk assessment and prevalence-based screening studies are necessary to evaluate and monitor the performance of syndromic management [1]. The aim of the present study was to document the pattern of common STIs and to evaluate the frequency of occurrence of STIs isolates among ARFH attendees in Ibadan, Southwestern Nigeria.

MATERIALS AND METHODS

Study Area: The study was carried out among attendees of Association for Reproductive Family and Health (AFRH) centre in Ibadan. Ibadan city lies 3°5' E and 7°23' N. The city is characterized by low level of environmental sanitation, poor housing and lack of potable water and improper management of wastes especially in the indigenous core areas characterized by high density and low income populations.

Study Population: Two hundred consecutive patients, who attended the STI clinic of a secondary health care centre (ARFH), with one or more of the complaints as enunciated by WHO in its syndromic approach for the diagnosis of STI [19] were included as subjects. All were screened for common STIs by standard microbiological methods [20]. Table 1 shows demographic profiles of the attendees of AFRH Clinic in Ibadan, Southwestern Nigeria.

Sample Collection: Samples of blood, urethral swab, high vaginal swab (HVS) and endocervical swab (ECS) were collected from males and females, respectively and subjected to direct examination by Gram staining and culture plate inoculation at the site of sample collection. Ten milliliters venous blood (without anticoagulant) was collected aseptically from all patients. Sera were separated and stored at-20°C in screw-capped glass tubes.

Table 1: Demographic profiles of the attendees of AFRH Clinic in Ibadan, Southwestern Nigeria

Profiles	No. Tested (%)	No. males (%)	No. females (%)
Age Group (years)			
16-29	98(49.0)	18(18.4)	80(81.6)
30 and above	102(51.0)	31(30.4)	71(69.6)
Sex			
Males	49(24.5)	49(100.0)	0(0.0)
Females	151(75.5)	0(0.0)	151(100.0)
Marital status			
Married	141(70.5)	15(10.6)	126(89.4)
Single	59(29.5)	34(57.6)	25(42.4)
Total	200(100.0)	49(24.5)	151(75.5)

Isolation and Identification of Isolates

***Neisseria gonorrhoeae*:** A presumptive diagnosis of gonococcal infection was made on observing polymorphonuclear leucocytes (PMNLs) with Gram-negative intracellular diplococci (ICDC). If the smear showed five or more PMNLs in the absence of Gram-negative ICDC, a presumptive diagnosis of nongonococcal urethritis (NGU) was made in men [21]. For the isolation of *Neisseriagonorrhoeae*, swabs were directly inoculated on the chocolate agar plate containing vancomycin, colistin and amphotericin-B and incubated in 5-10% carbon dioxide for 24-48 h. Isolates were identified as *N. gonorrhoeae* on the basis of colony morphology, Gram staining, oxidase test and rapid carbohydrate utilization test (RCUT) [22].

***Candida* Species:** For the isolation of *Candida*, urethral/cervical discharge was inoculated on Sabouraud dextrose agar and identification was done by standard mycological techniques [20].

***Gardnerella vaginalis*:** This was carried out according to the methods of Cheesbrough [22]. *Gardnerella vaginalis* was identified by a combination of Gram staining reaction and the pH of the discharge. The wet preparation showed abundance of 'clue cells' [squamous epithelial cells whose surfaces were smothered with masses of micro-organisms], the pH of the saline preparation was found to vary between 5.0-5.6 [i.e. higher than normal pH of 3.0-4.5] when measured with a pH indicator paper (BDH, UK) and in a Gram stain of positive cases, the normal lactobacilli flora was almost or completely replaced with masses of Gram variable organisms. Submitted specimens were inoculated onto enriched blood agar (EBA) (tryptic soy agar base [Difco, Detroit, Mich.] supplemented with 5%

defibrinated sheep blood [Cleveland Scientific, Bath, Ohio], 1% horse serum [BBL, Becton Dickinson, Cockeysville, Md.] and 1% yeast extract [GIBCO Laboratories, Lawrence, Mass.]), on phenyl ethanol agar (BBL) supplemented with 5% sheep blood and on MacConkey agar (Difco). Specimens were also cultured in thioglycolate supplemented with 1% hemin (Sigma Chemical Co., St. Louis, Mo.) and 1% vitamin K (Sigma). The EBA and phenyl ethanol agar plates were incubated for 96 h at 35 to 37°C in a 5% CO_2 environment. The MacConkey agar and thioglycolate were incubated aerobically at-5 to 37°C for 24 and 96 h, respectively. Nonhemolytic colonies appearing on EBA after 48 h of incubation were subcultured onto EBA for further characterization. A commercial bacterial identification system (Rapid STREP bacterial identification system; Analytab Products, Inc., Plainview, N.Y.) was used to identify gram-negative to gram-variable bacilli that were catalase and oxidase negative. Additional tests, which have been used as a means of identifying *G. vaginalis* included detection of hemolysis on media containing sheep, rabbit, or single-or bilayer human blood; susceptibility to metronidazole, sulfisoxazole and sodium polyanetholesulfonate (SPS). Enriched blood agar was used to detect hemolysis of sheep blood. Tryptic soy agar base (Difco) supplemented with 5% defibrinated rabbit blood (Cleveland Scientific), 1% yeast extract (BBL) and 1% horse serum (GIBCO) was used to detect hemolysis of rabbit blood. Human blood-Tween (HBT) agar (BBL) was used to detect diffuse beta-hemolysis on bilayer human blood agar. Vaginalis agar (BBL) was used to detect diffuse beta-hemolysis on single layer human blood agar.

Other Organisms: The swab samples collected were inoculated onto Mac Conkey agar and Sheep Blood agar and incubated at 35ºC-37ºC for 16-48 hours. The isolates were identified to species level by conventional biochemical tests as described by Cheesbrough [22].

Wet Mounts Preparation and Direct Examinations: Normal saline wet mount examinations were done to detect motile trophozoites of *Trichomonas vaginalis* and yeast cells for *Candida* infection. A direct smear was made from the ulcer, if any and subjected to direct examination by Gram staining and Leishman staining for the presence of multinucleated giant cells, shoals of fish bacilli, or safety pin-appearing bacilli to detect herpes simplex virus (HSV), *Hemophilus ducreyi* and *Calymmatobacterium granulomatis*, respectively [20].

Data Analysis: The proportions were calculated for various disease prevalences.

RESULTS

Of the two hundred (200) patients, 151 females and 49 males tested, 110 (55.0%) were infected with various STI agents and 21 (10.5%) had growth of *Staphylococcus aureus*, *E. coli*, coliforms and normal genital flora. Table 2 shows the incidence of laboratory-confirmed sexually transmitted infection.

Prevalence of Laboratory-Confirmed Sexually Transmitted Infection: The prevalence of various STIs based on laboratory tests has been shown in Table 2. It showed that *Candida albicans* accounted for the maximum number of STI [54(27.0%)], followed by *Gardenella vaginalis (Bacterial vaginosis)* [23(11.5%)] and *Trichomonas vaginalis* [3(1.5%)]. *Treponema pallidum* (syphilis), *Nesseria gonorrhoeae*, chlamydial infection and HCV were not detected. In all, 55.0% had more than one STI concomitantly at the time of presentation. Other isolates include *Staphylococcus aureus* 4(2.0%), *E. coli* 1(0.5%), coliforms 1(0.5%) and normal genital flora 15(7.5%).

Prevalence of Laboratory-Confirmed Sexually Transmitted Infection in Relation to Sex: The prevalence of various STIs in relation to sex of patients has been shown in 3. It showed that *C. albicans* was predominantly

Table 2: Laboratory diagnosis, prevalence of sexually transmitted infections pathogens

Isolate	Total (n= 200); No.* (%)
Candida albican	54(27.0)
Gardnerella vaginalis (Bacterial vaginosis)	21(10.5)
Trichomonas vaginalis	3(1.5)
Chlamydia trachomatis	0(0.0)
Trepanema palladium (Syphilis)	0(0.0)
Neisseria gonorrheae	0(0.0)
Total	78(39.0)
Other isolates	
Staphylococcus aureus	4 (2.0)
E. coli	1 (0.5)
Coliforms	1 (0.5)
Normal genital flora	15 (7.5)
Total	21 (10.5)

* = Multiple responses

Table 3: Laboratory diagnosis, prevalence of sexually transmitted infections pathogens in relation to sex of patients

Isolate	Total (n= 200); No.* (%)	Males (n=49); No.* (%)	Females (n=151); No.* (%)
Candida albican	54(27.0)	8(16.3)	46(30.5)
Gardnerella vaginalis	21(10.5)	0(0.0)	21(13.9)
Trichomonas vaginalis	3(1.5)	0(0.0)	3(1.9)
Total	78(39.0)	8(16.3)	70(46.4)

* = Multiple responses

Table 4: Laboratory diagnosis, prevalence of sexually transmitted infections pathogens in relation to ages of patients

Isolate	Total n= 200 No.* (%)	Age group 16-29 yrs (n=98);No.* (%)	Age group 30 yrs and above (n=102); No.* (%)
Candida albican	54(27.0)	31(31.6)	23(22.5)
Gardnerella vaginalis	21(10.5)	13(13.3)	8(7.8)
Trichomonas vaginalis	3(1.5)	0(0.0)	3(2.9)
Total	78(39.0)	44(44.9)	34(33.3)

* = Multiple responses

Table 5: Laboratory diagnosis, prevalence of sexually transmitted infections pathogens in relation to marital status of patients

Isolate	Total (n= 200) No.* (%)	Married (n=141); No.* (%)	Singles (n=59); No.* (%)
Candida albican	54(27.0)	25(46.3)	34(57.6)
Gardnerella vaginalis	21(10.5)	6(4.3)	15(25.4)
Trichomonas vaginalis	3(1.5)	1(0.7)	2(3.4)
Total	78(39.0)	32(22.7)	51(86.4)

* = Multiple responses

higher in females [46(30.5%)] than in male patients [8(16.3%)]. *G. vaginalis* [21(13.9%)] and *T. vaginalis* [3(1.9%)] was found in females only. There was a significant association (P<0.05) between sex and infection acquisition (Table 3).

Prevalence of Laboratory-Confirmed Sexually Transmitted Infection in Relation to Age: Table 4 shows the prevalence of laboratory-confirmed sexually transmitted infection in relation to the ages of the AFRH attendees in Ibadan, Southwestern Nigeria. The age specific distribution of STI pathogens among patients in the study shows that those in the age group of 16-29 years had a higher prevalence rate of 44(44.9%) with all of them testing positive to one or more pathogens except *T. vaginalis*. While those in age group 30 years and above had a lower prevalence rate of 34(33.3%) with all of them testing positive to one or more pathogens (Table 4). There was a significant association (P<0.05) between age groups and infection acquisition.

Prevalence of Laboratory-Confirmed Sexually Transmitted Infection in Relation to Marital Status: Table 5 shows the prevalence of laboratory-confirmed sexually transmitted infection in relation to the marital status of AFRH clinic attendees in Ibadan, Southwestern Nigeria. It showed the occurrence of infections among these patients by marital status. Infection rates among these patients showed that married patients [32(22.7%)] had lower infection rate compared to their single counterparts [51(86.4%)]. Statistically, marital status was significantly associated with infection occurrence (P<0.05).

DISCUSSION

There is a dearth of information regarding the epidemiology of STIs in most countries for many reasons such as stigma and discrimination associated with the STI, lack of interdepartmental coordination for studies, poor attendance of STI patients at the public clinics and academic institutions and availability of limited diagnostic facilities. This in-depth analysis offers an important insight into the burden and pattern of various STIs and on the performance of laboratory diagnosis. Available data show that sexually transmitted diseases constitute great medical, social and economic problems in Nigeria [23]. Apart from the heavy affliction of urban dwellers, there is rapid excursion of these diseases to the rural areas as well. This situation is serious enough to attract government attention so that necessary control measures may be initiated in good time in order to avert the serious consequences.

In this study, the overall prevalence rate for STIs was 39.0% (n=78). This is lower compared to what was previously reported by some other authors. In a study by Kehinde and Lawoyin [24], one-hundred-and-eighty (85.7%) had an STI, of which 41 (22.8%) were co-infected with HIV. Even though the prevalence of trichomoniasis and candidiasis are rather high, bacterial vaginosis is the leading cause of vaginitis and vaginal discharge in Nigeria [23]. The predominant malignancy of women in Nigeria is cervical cancer which may be due to the high rates of infection of trichomoniasis and Herpes virus II. In our study, the peak age group of subjects with STI ranges from 16 to 29 years [44(44.9%)] and vast majority of them were female [70(46.4%)], thus constituting the major bulk of the subjects with laboratory confirmed STI. This is a matter of concern in the context of HIV transmission as genital ulcer facilitates the transmission of and enhances susceptibility to HIV infection by sexual contact [1, 25-26] while nonulcerative STIs like gonorrhea and chlamydia increase shedding of the HIV virus in the genital tract by recruiting HIV-infected inflammatory cells as part of normal host response [1,25].

In the present study, *Candida albican* (27.0%) was the commonest infection followed by *G. vaginalis* (10.5%) and *T. vaginalis* (1.5%). A marked decline in bacterial STIs, resulting in an apparent increase in fungal and viral STIs, has been reported from different regions [1, 15-16, 27]. Many people believe they have an STD and do not, yet they insist they do. This phenomenon may be a result of the common fear of infertility which results from STDs [23]. The present study showed laboratory confirmed STIs which showed *Candida albican* as the most predominant pathogen.

Bacterial vaginosis (BV) is a clinical entity that is characterized by a change in vaginal ecology where the normal flora of *lactobacillus* morphotypes is replaced by a mixed microbial flora consisting of *anaerobes* and *Gardnerella vaginalis* [28-29]. *Gardnerella vaginalis* is a marker for a variety of infections caused by different bacteria including aerobic and anaerobic streptococci and staphylococci. The normal microbial flora of the vagina plays an important role in preventing genital and urinary tract infections in women [28]. This organism and other sexually transmitted pathogens are associated with high risk for HIV infection [30] by causing genital lesions which facilitate viral entry or by increasing the number of target cells for HIV (activated monocytes) [29, 31].

The present studyrevealed that the overall prevalence of *G. vaginalis* was 10.5%. This figure is lower than the 25.0% reported for *G. vaginalis* by Alli *et al*. [29]; the 86.0% prevalence rate reported by Fernández-Limia *et al*. [32]; the 68.0% and 40.0% reported by Oyewole *et al*. [33] among HIV-infected and non-infected women in Sagamu, Ogun state, Nigeria respectively; and the 26.0% reported by Nwadioha *et al*. [34] among women in Aminu Kano Teaching Hospital, Kano, Nigeria; the 28.0% reported by Shazia *et al*. [35] among women at Ayub Teaching Hospital, Abbottabad, Pakistan; the 26.05% reported by Rao *et al*. [36] among in a rural setup in India; and the 25% in Nepal [37]. However, the prevalence of *G. vaginalis* in this study was much higher than some other published reports [38-40]. It is higher than the 0.9% prevalence rate reported by Usanga *et al*. [7] among non-pregnant women in Calabar, Nigeria; and comparable to the 2.1% prevalence rate reported by Usanga *et al*. [7] among pregnant women in Calabar, Nigeria.

Also, *G. vaginalis* was found in females only; 21(13.9%) of the females in this study had *G. vaginalis* colonization. This is comparable to what has been earlier reported [29, 32]. According to Shazia *et al*. [35], Alteration in balance of normal vaginal organisms can cause the overgrowth of the bacteria that creates vaginal discharge. It is common among sexually active women yet there still remain gaps in our knowledge of this infectious disorder [29, 35].

Trichomonas vaginalis may be emerging as one of the most important cofactors in amplifying HIV transmission, particularly in African-American communities of the United States [41]. In a person co-infected with HIV, the pathology induced by *T. vaginalis* infection can increase HIV shedding; *Trichomonas* infection may also act to expand the portal of entry for HIV in an HIV-negative person [41]. Peak prevalence of *T. vaginalis* 2.9% occurred in the females, age group 30 years and above while none of the subjects below 30 years of age had trichomoniasis. This deviate from the findings of Adeoye and Akande [42], who reported that a peak prevalence of 1.8% in the age group 21-30 years while none of the subjects below 21 years and above 50 years had trichomoniasis. Nimorsi *et al*. [43] also reported the highest *Trichomonas vaginalis* infection in female subjects within 20-25 years old. Mason *et al*. [44] reported that after controlling for age, seropositivity was significantly associated with being sexually active, having multiple sex partners, having a partner who had multiple sex partners and having a new sex partner in the past year.

The incidence of trichomoniasis is highest in women with multiple partners and in groups with a high prevalence of other STIs [45]. On univariate analysis, there was an association, although not statistically

significant, between the existence of *T. vaginalis*, multiple sexual partners, drug addiction and no condom use [45]. A statistically significant association was found between trichomoniasis, prostitution and other STDs [46]. Barberis [47] reported the prevalence of mixed infections with *T. vaginalis* and *Gardrenella vaginalis* (5.5%) and with *Neisseria gonorrhea* (2.2%). Studies by Sorvillo [41] and Shuter [48] revealed that *T. vaginalis* might serve as co-factor in HIV transmission and also in amplifying its transmission particularly in African-American communities in the United States. In Nigeria, cases of *Trichomonas vaginalis* have been reported. Nimorsi *et al*. [43] recorded the occurrence of urinary schistosomiasis and trichomoniasis co-infection in the genito-urinary tract of 14 (6.3%) female inhabitants of Ikao Village, in Owan Local Government Area of Edo State, Nigeria. In a study by Adeoye and Akande [42], the overall prevalence of *T. vaginalis* was more prevalent (5.4%) at Military Hospital than at LUTH (1.9%).

CONCLUSION

The study confirmed a similar pattern of higher incidence of fungal STIs which could be due to the increased usage of antibiotics. Also, a high incidence of STI in the study population indicates the close association of STI isolates and the importance of early diagnosis of these curable diseases. Previous studies from different parts of the world have also supported these observations [1, 27]. In conclusion, fungal STIs constitute the major burden of the STI clinic and enhance the susceptibility of an individual to acquire or transmit HIV through sexual contact. The government should allocate adequate funds for health programs and research, particularly those associated with STDs. This study also shows a high STI rate indicating that there is need for proper management of STI, as this will help curb the spread of HIV infection in Nigeria.

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