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Full Length Research Paper

Prevalence of tuberculosis and human immunodeficiency virus (TB/HIV) co-infections amongst patients with bronchopulmonary disorders in Lagos

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Tuberculosis (TB) still remains a global public health problem of enormous dimension. This study was carried out between March 2007 and June 2008 to determine the prevalence of TB and TB/HIV co-infections amongst patients with bronchopulmonary disorders in Lagos State, Nigeria. 1280 patients were screened for acid-fast bacilli (AFB) by microscopy and culture at the TB reference laboratory of the Nigerian institute of medical research Lagos. Information on HIV status of each patient was obtained from medical records. Of the 1,280 patients, 318 (24.8%) were diagnosed positive for AFB while 236 patients (18.4%) were co-infected with TB and HIV. Amongst the co-infected patients, TB was diagnosed in 45 (14%) by microscopy alone, in 145 (46%) by culture alone and in 46 (15%) by both microscopy and culture. The higher yield of TB cases by culture alone compared to microscopy alone further confirms the higher sensitivity of culture for TB case detection amongst TB/HIV co-infected patients. In addition, the high burden (24.8%) of TB amongst the screened patients calls for differential diagnosis of this infection amongst cases with bronchopulmonary disorders seen in health facilities.

Key words: Tuberculosis, human immunodeficiency virus, TB/HIV co-infections.

INTRODUCTION

Tuberculosis remains a global health problem and is responsible for more deaths than any other infectious disease (Taura et al., 2008). It is estimated that about one billion individuals are infected world wide, with 10 million new cases and over 3 million deaths per year (Taura et al., 2008). However, the emergence of the human immunodeficiency virus has contributed to a global resurgence of tuberculosis. In sub-sahara Africa, the largest increase in new cases of tuberculosis has occurred in countries with the highest HIV prevalence rate (Richard and Chaisson, 2007), despite successful implementation of World Health Organization (WHO) recommended TB control strategies.

Individuals infected with TB have a higher likelihood of progressing from latent TB to active TB if they are co-infected with HIV (Ahmed et al., 2007). Furthermore, TB may speed the development of HIV infection to AIDS in dually infected individuals. The timely diagnosis of TB is therefore important in TB/HIV co-infection. Nevertheless, diagnosis of TB in these groups of individuals is becoming increasingly problematic as the yield from microscopy is becoming very low. As a result, most cases of HIV positive smear negative TB cases are missed in diagnosis (Thuy et al., 2007). The sensitivity of the

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Abbreviations: TB, Tuberculosis; **HIV,** human immunodeficiency virus; **DOTS,** directly observed treatment short-course; **AFB,** acid-fast bacilli; **WHO,** World Health Organization; **ARV,** antiretroviral; **LJ,** Lowestein Jensen; **ZN,** Ziehl Neelsen; **MGIT,** mycobacterial growth indicator tube.

sputum smear examination is reduced in HIV positive patients due to the lower rate of caseation necrosis and consequent lower number of acid-fast bacilli (AFB) in the airway (Matee et al., 2008) . Thus, the prevalence of smear negative tuberculosis is on the rise. This may delay diagnosis and treatment of TB, potentially leading to greater TB transmission in the community. Sputum culture for acid-fast bacilli is the gold standard and much more sensitive than the routine smear microscopy (Kagu and Ayilara, 2007). It can detect lesser number of AFB in sputum samples (10 - 100 viable bacilli per ml) for positive smear result (Wood, 2007). It has been shown that HIV-positive patients has higher prevalence of culture-positive TB and undiagnosed smear positive TB than HIV-negative individuals (Harries et al., 2010). However, culture is not readily available in most of the laboratories in developing countries due to cost and inability to meet up with the world health organization (WHO) stringent infrastructural requirements (Thuy et al., 2007).

In the past few years, Nigeria has witnessed a resurgence of new cases of TB and the growing number of new cases of HIV. Under the present setting there are bound to be cases of TB and HIV co-infections. Several reports on TB and TB/HIV co-infections are available in literature (Taura et al., 2008; Okogun et al., 2002; Idigbe et al., 1994; Umeh et al., 2007) with limited number emanating from Lagos. This study is therefore designed to study the prevalence of TB and TB/HIV co-infections in Lagos.

MATERIALS AND METHODS

Study population

The research work was carried out at the National Tuberculosis Reference Laboratory of Nigerian Institute of Medical Research (NIMR), Yaba, Lagos. Patients who presented with bronchopulmonary disorders at the NIMR DOTS clinic were used for the study. Most of the HIV positive patients were referred from ARV clinic of NIMR. Sampling was non- randomized as sputum samples were collected from all patients who visited the DOTS clinic. Inclusion criteria included patients aged 10 years and above with clinical symptoms such as abnormal chest X-Ray and cough for more than 3 weeks. Because of the inability of children below 10 years to produce quality sputum (Palomino et al., 2007) used in this study, they were excluded. All patients gave informed consent for study participation. Ethical approval for the study was received from institutional review board of Nigerian institute of medical research

Collection of samples

Three consecutive sputum samples (spot, early morning, spot) were collected from the patients aged 10 years and above in sterile wide-capped containers from March 2007 and June 2008. Prior to sputum collection, demographic information on each of the patients was obtained with a well structured questionnaire and corresponding medical records. Result of HIV test and confirmation were extracted from the patients medical records obtained from the clinical science laboratory of Nigerian institute of medical Research.

LABORATORY ANALYSIS

Microscopy

All the sputum specimens were analysed in a biosafety cabinet (BSC) for the presence of acid-fast bacilli using Ziehl Nelsen staining technique as described by WHO (1998). Briefly, the sputum samples were decontaminated using the modified Petroff's method (Kent and Kubica, 1985). Smears of the final deposits were made on clean grease free slides and allow to air dry. An air dried smear of concentrated sputum was covered with carbol fuchsin, steamed for 5 min and was decolourised with acid-alcohol for about 3 min after cooling. The slide was rinsed with water and counter stained with methylene blue for 1 min. The slide was rinsed with water, cleaned and allow to air dry. The stained slides were then examined under oil immersion using Olympus light microscope (model CX41) for acid fast bacilli.

Culture

About 2 - 3 drops of the final sputum deposits of the decontaminated sputum samples were evenly spread on the entire surface of Lowestein-Jensen (LJ) slopes and subsequently incubated at 37°C. The slopes were examined for growth on day 3, 7 and weekly thereafter for 8 weeks and subsequently on day 63 after inoculation. All slopes not showing any growth within this period were discarded and recorded as AFB negative. For slopes with growths, the resultant colonies were examined for morphology pigmentation and acid-fastness. Smear of colonies were made, stained by Ziehl-Neelsen (ZN) technique and examined for the presence of acid-fast bacilli. Isolates that were acid-fast bacilli were confirmed as positive.

Statistical analysis

Analysis of data in this study was carried out using the SPSS for windows version 15.0.

RESULTS

Between March 2007 and June 2008, 1280 patients with bronchopulmonary disorders were studied. The sex and age distribution are shown in Table 1. There are slightly more females (54.1%) than males (45.9%). For both sexes, 3% were in the age group 10 - 20 years, while majority of the patients (89.9%) were within the age group 21 - 50 years and 7.1% belong to the age group 51 and above.

The sex and HIV status of the patients are shown in Table 2. Analysis of data generated from the clinical records of the patients showed that 1037 (81%) of the 1280 patients were positive for HIV while 243 (19 %) were negative.

In this study, a patient was diagnosed as a case of tuberculosis when one or all the sputum samples were positive for AFB by microscopy, culture or both. The age and sex distribution of the TB positive cases are shown in Table 3. Of the 1280 patients screened 318 (24.8%) were positive for AFB. Among these 318 confirmed cases of TB 145 (45.6%) and 173 (54.4%) were male and female

Table 1. Age and sex distribution of the patients with bronchopulmonary disorder.

Age group (yrs)	Male (%)	Female (%)	Total
10 - 20	23(1.8)	15 (1.2)	38(3.0)
21 - 30	106 (8.3)	285 (22.3)	391(30.6)
31 - 40	250(19.5)	252 (19.7)	502(39.2)
41 - 50	149(11.6)	108 (8.4)	257(20.1)
51 - 60	46(3.6)	21 (1.6)	67(5.2)
>60	13(1.0)	12 (0.9)	25(1.9)
Total	587(45.9)	693 (54.1)	1280 (100)

Yrs = Years, % = percentage, > =greater than.

Table 2. Sex and HIV status of the patients with bronchopulmonary disorder.

Sex	HIV +ve (%)	HIV -ve (%)	Total
Male	449 (35.1)	138 (10.8)	587 (45.9)
Female	588 (45.9)	105 (8.2)	693 (54.1)
Total	1037 (81)	243 (19)	1280 (100)

% = percentage, HIV = human immunodeficiency virus, +ve = Positive, -ve = Negative.

Table 3. Age and sex distribution of TB positive patients.

Age (Yrs)	Number screened	Male (%)	Female (%)	Total
10 – 20	38	7 (18.4)	4 (10.5)	11 (28.9)
21 – 30	391	28 (7.1)	84 (21.5)	112 (28.6)
31 – 40	502	56 (11.2)	59 (11.7)	115 (22.9)
41 – 50	257	34 (13.2)	20 (7.8)	54 (21.0)
51 – 60	67	17 (25.4)	5 (7.4)	22 (32.8)
>60	25	3 (12.0)	1 (4.0)	4 (16.0)
Total	1280	145 (11.3)	173 (13.5)	318 (24.8)

TB = tuberculosis, Yrs = Years, > = greater than, % = percentage.

respectively, ($p < 0.05$). The age group 21 - 40 years had the highest TB cases while the age group 61 years and above had the lowest TB cases.

The age and sex distribution of TB/HIV co-infected cases are shown in Table 4. It was observed that 236 of the study population had TB and HIV co- infection and this gave a prevalence rate of 18.4%. Of all the patients diagnosed of TB, 74% (236/318) were HIV positive. The distribution showed that among this 236 co-infected cases, 99 were male and 137 were female, ($p < 0.05$). Also the age group 21 - 40 years had the highest cases of TB/HIV co-infection while the age groups 10 - 20 years and above 60 years had the lowest TB/HIV cases. Correlation between the result of microscopy and culture for the diagnosis of acid-fast bacilli in TB/HIV and TB/non- HIV patients is shown in Table 5. Among the positive cases 175 (55%) were detected by culture alone, 61 (19%) were detected by microscopy alone while 82 (26%) were detected by both culture and microscopy. The difference between the detection for culture and microscopy was statistically significant ($p < 0.05$).

DISCUSSION

In the recent decades, the number of TB cases has increased by several folds especially in sub-African countries. In Nigeria, several studies have examined prevalence of TB among Nigerian population (Taura et al., 2008; Umeh et al., 2007; Nwachukwu et al., 2009) . In the studies, prevalence of TB ranges from 15.7 and 23.8% between the ages 10 years and above. From our study, prevalence rate of 24.8% was recorded among patients with bronchopulmonary disorders seen in Lagos which was slightly higher than the available data.

With regards to gender and TB positivity among these patients, the rate was statistically higher in females than in males. This is in contrast with the studies of Taura et al., 2008 in Kano; Umeh et al., 2007 in Nasarawa and Nwachukwu et al., 2009 in Abia. The differences in the infection rate in females and males could be as a result of biological factors such as higher susceptibility to infection due to low immunity in women. Furthermore, in both sexes, the age group 21 - 40 years had higher TB rate than the other age group. This observation is in line with the studies by other researchers (Nwobu et al., 2004; Umeh et al., 2007; Nwachukwu et al., 2009) and further confirms that these age groups are more vulnerable to TB infection.

The growing association between TB and HIV is globally recognized (Ahmed et al., 2007; Thuy et al., 2007; Nsubuga et al., 2002). Similar reports are found in Nigeria (Okogun et al., 2002; Idigbe et al., 1994; Umeh et al., 2007). A co-infection rate of 18.4% was recorded in this study and this was higher than the rates in earlier studies. This variation in co-infection rates could be linked to the fact that majority (81%) of the patients screened were referred from antiretroviral (ARV) care and treatment clinics. This study has further elaborated previous findings. There was a high prevalence of TB (74%) in HIV positive patients. This may be due to the fact that majority of study population were referrals from ART center. Our study is in line with that of Kwanjana et al. (2001) supporting the opinion on high rates of TB/HIV co-infections in high HIV prevalence population. The co-infection was higher in females than in males. The difference in co-infection rate in males and females could be as a result of early exposure of females to sexual activity due to bad economic situations (Umeh et al., 2007) , high susceptibility to infection (Umeh et al., 2007), delay in care seeking due to stigma associated with HIV infection (Nsubuga et al., 2002); less access to fund for transportation and personal health care.

Table 4. Age and sex distribution of TB/HIV co-infected patients.

Age (Yrs)	Number screened	Male (%)	Female (%)	Total
10 – 20	38	1(2.6)	3(7.9)	4 (10.5)
21 – 30	391	14 (3.6)	65(16.6)	79 (20.2)
31 – 40	502	40 (8.0)	50 (9.9)	90 (17.9)
41 – 50	257	26(10.1)	16 (6.2)	42 (16.3)
51 – 60	67	16(23.9)	2(3.0)	18 (26.9)
>60	25	2(8.0)	1(4.0)	3 (12.0)
Total	1280	99 (7.7)	137 (10.7)	236 (18.4)

TB = tuberculosis, HIV = human immunodeficiency virus, Yrs = Years, > = greater than, % = percentage.

Table 5. Correlation of results of microscopy and culture for acid-fast bacilli in TB/HIV and TB/non-HIV patients.

Method and Result	TB/HIV (%)	TB/non-HIV (%)	Total (%)
Microscopy negative + culture positive	145 (46)	30 (9)	175 (55)
Microscopy positive + culture negative	45 (14)	16 (5)	61(19)
Microscopy positive + culture positive	46 (15)	36 (11)	82(26)
Total positivity	236 (75)	82 (25)	318(100)

TB = tuberculosis, HIV = human immunodeficiency virus, % = percentage.

Diagnostic technique for TB screening is centered on microscopy and culture on liquid or solid media. In this study, the patients were screened for AFB by both microscopy and culture. It was observed that more AFB was detected by culture than microscopy and this is statistically significant ($p < 0.05$). Furthermore, correlation of the AFB result by microscopy and culture in TB/HIV co-infected patients and TB/non-HIV patients shows that culture detected more AFB than microscopy. This data corresponds with observations recorded in earlier reports (Kagu and Ayilara, 2007; Aderaye et al., 2007) and further confirmed the decreased sensitivity of smear microscopy for the diagnosis of TB. The implication indicated that 36% were missed by microscopy but picked up by culture. The consequences for this low sensitivity of microscopy are several including delayed or misdiagnosed cases, contributing to delay in treatment and increased morbidity and mortality rates (Kagu and Ayilara, 2007). Culture of *Mycobacterium tuberculosis* remains the gold standard for TB diagnosis because it is sensitive and specific for TB in both HIV-positive and HIV-negative individuals (Wood, 2007).

Hence, these findings have indicated an urgent need for the country to increase capacity for culture facilities in TB laboratories. Recent techniques are centered on the use of rapid methods such as Bactec MGIT 960, Hain assay and line probe assay in the diagnosis of TB infection. Also, due to high TB prevalence among HIV patients in this study, we recommend that all HIV patients be screened for TB likewise, all TB patients be screened for HIV.

In conclusion, given the high rate of TB and TB/HIV co-infections, this study highlighted the need for differential diagnosis of TB among cases of bronchopulmonary disorder seen in health facilities. Majority of cases with co-infections were detected by culture, there is therefore need to emphasize the use of culture for active case finding of TB especially in HIV positive patients.

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