Background: Tuberculosis (TB) remains a global public health problem and one of the top ten leading causes of death, worldwide, with developing countries bearing the highest burden. Nigeria is first in Africa and sixth in the world among the countries with the highest TB burden, but is among the 10 countries accounting for over 70% of the global gap in TB case detection and notification. Enugu State, Nigeria reportedly has a notification gap of almost 14,000 TB cases; a situation which must be addressed.

Materials and Methods: A total number of 868 individuals accessing DOTS services in designated centres within the six Local Government Areas (LGAs) of Enugu North geographical zone, was recruited into the study. The participants were screened for HIV seropositivity by standard protocols, while screening for TB and drug-resistant TB were conducted by a combination of Zhiel Neelsen staining and Nucleic Acid Amplification Test (Xpert® MTB/Rif).

Results: Of the 868 subjects that participated in the study, 176 (20.3%) were HIV seropositive. The highest prevalence (26.7%) of HIV was recorded in Udenu LGA, while the least (13.1%) was recorded in Nsukka LGA. Overall TB prevalence was found to be 22.1% and 21.3% by sputum-smear and NAAT, respectively. Uzo Uwani LGA recorded the highest prevalence of both TB (33.3%) and TB/HIV co-infection (16.7%), but the lowest prevalence of resistant TB. Nsukka LGA had the highest prevalence of resistant TB.

Conclusion: Enugu North geographical zone, Nigeria, has a high prevalence of both HIV and TB, including resistant TB and there is need to increase monitoring of individuals resident in this region.

Keywords: Mycobacterium tuberculosis, Nigeria, MDRTB, Enugu Nigeria, HIV, Xpert® MTB/Rif

List of Abbreviations: TB – Tuberculosis, MDRTB – Multidrug-Resistant Tuberculosis, WHO – World Health Organization, HIV – Human Immunodeficiency Virus, AIDS – Acquired Immunodeficiency Syndrome, NTBLCP – National Tuberculosis and Leprosy Control Programme, LGA – Local Government Area, NPC – National Population Commission, DOTS – Directly Observed Treatment Short-course, NAAT – Nucleic Acid Amplification Test

Introduction

Tuberculosis (TB) remains a global public health problem and one of the top ten leading causes of death, worldwide, with developing countries bearing the highest burden (WHO, 2020). In 2018, Nigeria was listed as first in Africa and sixth among the 30 countries of the world with the highest TB burden (WHO, 2020). Unfortunately, the problem of TB in Nigeria has been complicated by the emergence and spread of drug resistant TB and a high burden of HIV/AIDS (NTBLCP, 2017; WHO, 2018). The problem of TB is worsened when there is also a high burden of HIV infections, as people with HIV are more likely to develop active TB. According to WHO reports, an estimated 63,000 Nigerians living with HIV/AIDS develop TB, while about 39,000 die from the disease, each year (WHO, 2018).

To further compound the problem, Nigeria is ranked among the 10 countries that accounted for 77% of the global gap in TB case detection and notification in 2016. It is reported that Nigeria contributes about 8% of the 4.3 million TB cases missed globally.

Consequently, in 2019, WHO initiated ‘community-informant’ methods to find missing TB cases in Nigerian communities (WHO, 2019). The initiative covered 12 high burden states in Nigeria, but Enugu State was not part of the study. However, according to NTBLCP (2019), Enugu State has a notification gap of almost 14,000 TB cases.

This study was conducted in Enugu North geographical zone. The zone comprises six Local Government Areas (LGAs), namely,Nsukka, Igbo-Etiti, Uzo-Uwani, Igbo Eze North, Udenu and Igbo Eze South, and had a total ward population of about 1,466,996 at last count (NPC, 2012). The zone is characterized by low socio-economic indicators and a majority of the residents engage in low-
income generating activities, such as petty trading and peasant farming. In addition, one of the commercial nerve-centres of the zone, Obollo Afor, in Udenu LGA, is also a stopover for commercial drivers and people on transit to and from the northern parts of the country, making the area a hub for commercial sex activity, with attendant problems, such as HIV transmission. The study was, therefore, conducted to determine the prevalence of TB and MDR-TB in the six different LGAs of Enugu north geographical zone, with a view to fill some of the gap currently existing in TB detection and reporting in Nigeria.

**Materials and Methods**

**Study area**

The study was conducted in Enugu north geographical zone, south eastern Nigeria, which currently constitute Enugu north senatorial zone in the state geopolitical arrangement and is comprised of six local government areas (LGAs). The zone has a total ward population of about 1,466,996 (NPC, 2012). The zone is characterized by low socio-economic indicator, with a majority of the residents engaging in low-income generating activities, such as small trading and peasant farming. The geographical zone has about 35 DOTS centres spread across the six LGAs. Sputum samples were collected from five comprehensive HIV/AIDS treatment sites (2 public health facilities and 3 faith-based organizations) and the other DOTS/ TB Microscopy centres in the zone (Figure 1).

![Figure 1: Map of Enugu North Geographical Zone Showing the DOTS Centres.](image)

**Study population**

The study group comprised 868 individuals of different age groups (from age 5 and above) accessing DOTS services in the designated centres across the 6 LGAs in the zone; with productive cough or presumptive diagnosis of tuberculosis, who volunteered to participate in the study. The survey population for NAAT and drug resistant TB comprised 207 stratified randomly selected individuals with productive cough, accessing clinical services in the 35 DOTS centres. The DOTS centres provided the opportunity to collect sputum samples from individuals with high suspicion index for tuberculosis, those newly diagnosed and those on treatment but on follow up visit to the facilities. Individuals less than 5 years and/or ≥ 5 years who clinically/symptomatically screened positive to tuberculosis but did not have productive cough were excluded from the study.

**Ethical consideration**

The study was carried out in accordance with the tenets of the declaration of Helsinki. The purpose of this study was explained to the subjects in English and Igbo languages. Code numbers rather than names were used to identify them to ensure confidentiality. An Informed Consent Form was developed, explained and administered to clients/clients’ care givers (for children from 5 and above). Only consenting clients were enrolled in the study. Participation in the study was completely voluntary and at no cost to the consenting clients. Clients were also informed that quality of care in the health facility would not in any way, be affected by their decision to participate or not. Approval was obtained from the authorities of district hospitals and the respective DOTS/TB microscopy centres in the zone. Ethical clearance was obtained from the Health Research and Ethics Committee of Enugu State Ministry of Health, Enugu (Ref. No.: MH/MSD/EC/0183).

**Diagnosis of HIV infection**

HIV testing was performed on all participants according to the national guidelines for HIV testing, using Determine, Unigold and Stat Pak rapid diagnostic test kits, according to the manufacturers’ instructions. The Determine kit was used as the first line strip for
HIV testing, according to the national guidelines. Positive results were retested with the Unigold kit. However, in the event of a discordant result, Stat Pak was used. The final result of the algorithm for a particular sample was scored positive if at least two of the three rapid screening HIV assays were reactive. HIV counseling was offered to all subjects with unknown HIV status and those who previously tested negative.

Collection and processing of sputum samples

The subjects were advised to rinse their mouth at least twice with water before producing the specimen in order to remove food and reduce contaminating bacteria load from their mouths. They were instructed to take two breaths, and then cough deeply and expectorate the sputum into provided 50 ml screw-capped translucent bottles, holding the sample container close to their mouth. Three sputum specimens (spot, early morning, spot) were collected from each participant under close supervision. The samples were then processed for the standard acid-fast direct smear microscopy using Ziehl-Neelsen staining and one of the spot samples processed for Rifampicin resistance (MDRTB) assay by Nucleic Acid Amplification Test (NAAT) at Enugu Ezike District Hospital TB laboratory, according to the National guidelines. Stained smears were examined for acid-fast bacilli (AFB) under the microscope using oil immersion (100X) objectives.

Confirmation of TB and drug-resistant TB

Confirmation of the presence of *M. tuberculosis* and resistance (MDRTB) were assayed by Nucleic Acid Amplification Test (NAAT), using the GeneXpert/MTB/RIF Kit, in a semi-nested rt-PCR reaction. The tests were run following the manufacturer’s instructions.

Results

Local Government and gender distribution of participants

A total number of 868 individuals was enrolled in the study comprising 420 (48.4%) males and 448 (51.6%) females. Of the 868 subjects from the six LGAs in the zone, Igbo Eze North had the highest number of participants, with 425 (48.9%) and Igbo Etiti had the least number of participants, with 29 (3.3%), as shown in Table 1.

| LGA          | Number of DOTS Centres | Number of Males (%) | Number of Females (%) | Total (%) |
|--------------|------------------------|---------------------|-----------------------|-----------|
| Igbo Etiti   | 5                      | 18 (2.1)            | 11 (1.3)              | 29 (3.3)  |
| Igbo Eze North | 5              | 187 (21.5)          | 238 (27.4)            | 425 (48.9) |
| Igbo Eze South | 5               | 63 (7.3)            | 65 (7.5)              | 128 (14.8) |
| Nsukka      | 8                      | 50 (5.8)            | 34 (3.9)              | 84 (9.7)  |
| Udenu       | 5                      | 81 (9.3)            | 91 (10.5)             | 172 (19.8) |
| Uzo Uwani   | 7                      | 21 (2.4)            | 9 (1.0)               | 30 (3.4)  |
| Total       | 35                     | 420 (48.4)          | 448 (51.6)            | 868       |

HIV seropositivity amongst participants from different LGAs

Out of 868 subjects that participated in the study, 176 (20.3%) were HIV seropositive. Of these, 97 (54.5%) were females and 79 (45.5%) males. The difference between male and female seropositivity was however not statistically significant (P < 0.05). Local government disaggregation of data on HIV seropositivity in both male and female participants showed that the highest prevalence (26.7%) was recorded in Udenu LGA, while the least (13.1%) was recorded in Nsukka LGA. The highest prevalence in males (30.0%) was found in Igbo Eze South and the least (9.5%) in Uzo Uwani LGA, while for females, the highest prevalence (33.3%) was found in Uzo Uwani and the least (11.6%) in Nsukka LGA (Table 2).

| LGA          | Number Examine | Number Positive (%) | Number Examine | Number Positive (%) | Total Number Positive/LGA (%) |
|--------------|----------------|---------------------|----------------|---------------------|-----------------------------|
| Male Subjects |               |                     | Female Subjects |                     |                             |
| Igbo Etiti   | 18            | 4 (22.2)            | 11             | 2 (18.2)            | 6 (20.7)                    |
| Igbo Eze North | 187        | 28 (14.9)           | 238            | 48 (20.2)           | 76 (17.8)                   |
| Igbo Eze South | 63            | 19 (30.0)           | 65             | 13 (20.0)           | 32 (25.0)                   |
| Nsukka      | 50            | 7 (14.0)            | 34             | 4 (11.6)            | 11 (13.1)                   |
| Udenu       | 81            | 19 (23.5)           | 91             | 27 (29.7)           | 46 (26.7)                   |
| Uzo Uwani   | 21            | 2 (9.5)             | 9              | 3(33.3)             | 5 (16.7)                    |
| Total       | 420           | 79 (18.8)           | 448            | 97 (21.7)           | 176 (20.3)                  |
Prevalence of pulmonary TB in different LGAs in the zone

Of the 868 subjects that participated in the study, 192 (22.1%) tested positive to Ziehl Neelsen staining technique with an overall male to female preponderance of approximately 3:2. The male to female preponderance of sputum smear positivity was consistent in all the LGAs sampled except for Uzo Uwani LGA, where there was female to male ratio of 55.6% to 25.9%. Analysis of the result showed that there was a significant difference (P < 0.05) between the prevalence of sputum smear positivity in male and female subjects that participated in the study. In terms of case detection in LGAs, Uzo Uwani recorded the highest prevalence of sputum smear positivity (33.3%), while Igbo Etiti had the least (13.8%) as shown in Table 3.

Table 3: Occurrence of Pulmonary TB in different LGA

| LGA          | Male Subjects | Female Subjects |                |                |                | Total Number Positive/LGA (%) |
|--------------|---------------|-----------------|---------------|---------------|---------------|-----------------------------|
|              | Number Examined | Number Positive (%) | Number Examined | Number Positive (%) | Number Examined | Number Positive (%) |
| Igbo Etiti   | 18            | 3 (16.7)         | 11            | 1 (9.0)        | 4 (13.8)       |                             |
| Igbo Eze North | 187         | 47 (25.1)        | 238           | 41 (17.2)      | 88 (20.7)      |                             |
| Igbo Eze South | 63           | 20 (31.7)        | 65            | 8 (12.3)       | 28 (21.9)      |                             |
| Nsukka       | 50            | 14 (28.0)        | 34            | 7 (20.6)       | 21 (25.0)      |                             |
| Udenu        | 81            | 21 (25.9)        | 91            | 20 (21.9)      | 41 (23.8)      |                             |
| Uzo Uwani    | 21            | 5 (23.8)         | 9             | 5 (55.6)       | 10 (33.3)      |                             |
| Total        | 420           | 110 (26.2)       | 448           | 82 (18.3)      | 192 (22.1)     |                             |

TB and HIV co-infection

Among the 868 subjects included in this study, 66 (7.6%) had the double burden of HIV and TB. Local Government distribution of the co-infections of HIV/TB showed that Uzo Uwani LGA had the highest prevalence of 16.7%, followed by Igbo Etiti LGA (10.3%), while Igbo Eze North LGA had the least prevalence (5.6%), as shown in Figure 2.

Figure 2: Prevalence of TB/HIV co-infection in different LGAs of Enugu North geographical zone

Report of TB prevalence by gender and age group

The highest prevalence of TB (34%) was found among male subjects in the age group of 35 – 44 years while that of female (27%) was in the age group 25 – 34 years. Amongst the young adults and middle aged, ranging from 25 to 64 years, sputum smear positivity was recorded more in male subjects within the different age brackets. However, amongst children and adolescents (5 – 24 years), results showed a female predominance of the sputum smear positivity (Figure 3).
Prevalence of MDRTB in different LGAs

Of the 207 sputum samples subjected to NAAT (Xpert® MTB/Rif test), 44 (21.3%) were positive. Of these, 6 (13.6%) were resistant. Nsukka LGA had the highest prevalence of resistant TB (30.0%), followed by Udenu LGA (16.7%), while Igbo Etiti and Uzo Uwani LGAs had the least prevalence with no resistance (Table 4).

Table 4: Prevalence of resistant TB in different LGAs.

| LGA          | NAAT Positive Samples | Number Resistant to RIF | % RIF Resistance |
|--------------|-----------------------|-------------------------|------------------|
| Igbo Etiti   | 3                     | 0                       | 0                |
| Igbo Eze North | 12                  | 1                       | 8.3              |
| Igbo Eze South | 11                  | 1                       | 9.1              |
| Nsukka       | 10                    | 3                       | 30.0             |
| Udenu        | 6                     | 1                       | 16.7             |
| Uzo Uwani    | 2                     | 0                       | 0                |
| Total        | 44                    | 6                       | 13.6             |

Discussion

The problem of TB globally has been found to be worsened by HIV/AIDS, as people with HIV have a much higher risk of developing active TB. According to health reports, about 45% of HIV-negative people with TB and nearly all HIV-positive people with TB, have a high risk of death, without appropriate treatment (WHO, 2020). HIV and TB, co-infection has been described as a lethal combination, as each disease speeds up the other’s progress; a realization which probably led to updating of the WHO TB/HIV treatment approach, in 2012, to place persons with HIV on TB-preventive therapy upon confirmation of HIV status, regardless of CD4 count (WHO, 2020). This makes it imperative that such cases be detected promptly and given appropriate treatment.

In this study, 868 subjects with cough, accessing treatment in DOTS centres within Enugu North geographical zone were screened for HIV seropositivity and TB. The prevalence of HIV among the subjects was 20.3%. The highest prevalence (26.7%) was recorded in Udenu LGA, while the least (13.1%) was in Nsukka LGA. Within the zone, Nsukka LGA represents the most cosmopolitan/urban area, within which a tertiary institution is located. The low prevalence of HIV in Nsukka LGA could be attributed to improved lifestyle and better enlightenment of the inhabitants. On the other hand, the high prevalence in Udenu LGA, though inconsistent with the reports of Okoli et al., (2013), was not surprising, since the LGA houses one of the commercial nerve-centres of the zone, Obollo Afor, which is believed to have a high risk of HIV transmission due to increased number of commercial sex workers.

The overall prevalence of TB among participants was about 22% and there was a male to female preponderance of approximately 3:2, that was consistent in almost all the LGAs. This prevalence was higher than WHO projected national prevalence (WHO, 2018). Although the study was a hospital-based analysis (considering that most of the subjects were accessing chest clinics) and the findings may not be generalized in terms of prevalence of TB, the trends are nevertheless important. The higher male to female infection ratio, is in agreement with WHO reports, which state that globally, men are significantly more at risk of contracting and dying from TB than women. In 2017 alone, close to 6 million adult men contracted TB and around 840,000 died from it, compared to an
estimated 3.2 million adult women who fell ill and almost half a million who died from the disease (Balasubramanian et al., 2004; WHO 2018).

In this study, although Udenu LGA had the highest prevalence of HIV, the highest TB prevalence (33.3%) was recorded in Uzo Uwani LGA, despite contributing only 3.4% of the total participants. In addition, the LGA also recorded the highest HIV/TB co-infection, suggesting a strong link between the TB infections and HIV, for this LGA. Moreover, this is the only LGA where there was a significantly (P<0.05) higher female to male ratio of infection, for both HIV and TB. This trend is potentially attributable to sociocultural factors. The LGA has a large population of women and widows, as there is a high death rate among the males. Furthermore, women in the area are known to frequently indulge in extra-marital affairs or prostitution for augmentation of income, after which they undergo a cleansing ritual, before returning to their husbands. This predisposes the women to high rates of HIV and TB infection. The overall prevalence of HIV/TB co-infection in this study was moderately high at 7.6%, while the prevalence in the different LGAs ranges from 5.6% to 16.7%. The findings are in line with previous studies conducted in Ethiopia (7.5%), Nigeria (7.8%) and Tanzania (8.5%) (Ngowi et al., 2008; Iliyasu and Babashani, 2009; Wondimeneh et al., 2012), but lower compared to 32.8% and 36.3% reported in some studies from Nigeria (Awoyemi, 2002; Egbe et al., 2016). Analysis of TB prevalence, by gender and age-group also supports a strong link between TB and HIV in Enugu North geographical zone, as the TB cases were highest in the age groups, which are more active and have higher HIV risks, in both males and females. Age disaggregation showed that 34% of the overall sputum smear positive TB was found among male subjects aged 35–44 years while highest prevalence (27%) occurred in females aged 25 – 34 years.

In this study, the prevalence of drug-resistant TB was 13.6%. This finding is lower than the report of a study in southern Nigeria, with prevalence 23.4% (Adejumo et al., 2018) and another from northern Nigeria with prevalence of 14.3% (Fadeyi et al., 2017). However, the prevalence is relatively high in comparison with reports of studies from other parts of Nigeria and other African counties such as Ethiopia and Burundi (Sanders et al., 2006; Osman et al., 2012; Adane et al., 2015; Egbe et al., 2016; Kuyinu et al., 2018). The variation in reported prevalence even within the same country may reflect the variations in sample size, access to health care facilities, and effectiveness of TB control programmes. A somewhat surprising finding from this study was Udenu LGA having the highest prevalence for HIV, while Uzo Uwani had the highest TB prevalence, and the highest prevalence of drug-resistant TB was in Nsukka LGA. This can, however, be attributed to the higher number of cases in male subjects, who are of course less likely to visit the hospital for care and more likely not to follow through with treatment, as suggested by other reports (Auld et al., 2014; Bor et al., 2015).

Non-completion of treatment has been the top factor among factors contributing to TB drug-resistance (WHO, 2020). It can be concluded from this study that Enugu North geographical zone, Nigeria, has a high prevalence of both HIV and TB, including multidrug resistant TB and there is need to increase monitoring of this region.

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