Prevalence of Rifampicin Resistance among Presumptive Pulmonary Tuberculosis Patients within Lagos and its Environs in South-Western Nigeria

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Abstract: Drug resistance (DR) is a major global health concern and currently implicated in fuelling the burden of multi-drug resistant tuberculosis (MDR-TB) in Nigeria. Overall, DR poses serious public health threat to TB control programmes particularly in TB endemic countries with limited resources. However, early and rapid detection of rifampicin resistance (RR), a surrogate marker for MDR-TB is important to reduce treatment period and transmission; with the overall goal of reducing the burden of the disease. The study successfully determined the prevalence of RR *Mycobacterium tuberculosis* (MTB) among presumptive pulmonary TB patients in Lagos and its environs. A retrospective study involving 1,453 TB patients was conducted using data extracted from the clinical register of presumptive TB patients screened for MTB and RR-TB using Xpert MTB/RIF assay at the Centre for Tuberculosis Research, Nigerian Institute of Medical Research (NIMR) between January, 2018 and August, 2019. The data was analysed using Statistical Package for Social Sciences (SPSS) version 23. Result shows that the overall prevalence of TB was 15.3% (222/1453). MTB infection was detected in 79 locations out of which eight had a high prevalence (15.7%-26.1%) of the disease. Notably, from the patients, RR-TB was 5.9% (13/222) among all TB confirmed cases, with four (30.8%) being females and those infected ranging from 22 to 75 years (34.85±15.01) years; with one person (male) being co-infected with HIV. This study highlights the prevalence of 5.9% rifampicin-resistance among pulmonary TB patients in the urban and peri-urban areas of Lagos, South-Western Nigeria.

Keywords: Prevalence, Rifampicin-resistance, *Mycobacterium tuberculosis*, GeneXpert.

INTRODUCTION

The emergence and transmission of multidrug-resistant tuberculosis (MDR-TB) has over the years posed a great challenge to global health; thereby complicating diagnosis, treatment and control of TB (WHO/IUATLD, 2008). The Global TB Report of 2016 estimated newly diagnosed and previously treated TB cases with MDR-TB at 3.9% and 21% respectively. In 2015, approximately 580,000 TB cases were resistant to at least rifampicin (RR-TB) globally and 480,000 of the population were resistant to both rifampicin and isoniazid (MDR-TB) with 250,000 deaths occurring due to MDR-TB/RR-TB in the same year. (WHO, 2015). On the average, 4.1% of newly diagnosed and 19% previously treated TB patients are estimated to be infected with MDR-TB worldwide in 2017 (WHO, 2017). Treatment of TB (Self-medication) without proper diagnosis and drug susceptibility testing which is a common practice in developing countries has increased the transmission of drug resistant (DR) strains (Arega *et al.*, 2019). Drug resistant-TB is a major cause of concern globally, and currently implicated in fuelling the burden of TB in Nigeria. Overall, this poses serious public health threat to TB control programmes particularly in TB endemic countries with limited resources. The low sensitivity and time-consuming nature of conventional diagnostic techniques such as direct microscopy and culture increase the need for more efficient diagnostic methods (Onyedum, 2017). Although culture and drug susceptibility testing are gold standards for diagnosis, they are time consuming.

Therefore, newer and more rapid methods of diagnosis of rifampicin resistance (RR) using molecular techniques such as Gene-Xpert assay has been adopted for use (Prasad *et al.,* 2018). This is important to reduce treatment period and transmission; consequently decreasing the burden of the disease.

The World Health Organisation (WHO) endorsed the Xpert MTB/RIF assay, which is a rapid and automated molecular system that detects both *M. tuberculosis* DNA and rifampicin-resistance (RR) associated mutations simultaneously. Research recognized that RR can be a surrogate marker for MDR-TB in more than 90% of the cases (Riordan *et al*., 2008). Hence, WHO recommends that RR-TB patients should be treated like patients with MDR-TB (WHO, 2016). Initially, this method was indicated for patients with TB/HIV co-infection, presumptive MDR-TB and paediatric TB patients (WHO, 2011). However, three years after its implementation, it was recommended for all TB suspected patients (WHO, 2016). Notably till date, there are few reports of RR-TB prevalence study among pulmonary TB patients in Nigeria. Recent studies by Audu *et al*. (2017) reported RR-TB prevalence of 12.1% in all TB diagnosed patients in Nassarawa State while Adejumo *et al*. (2018) reported a higher prevalence of 22.5% among newly diagnosed TB patients in Lagos State.

Lagos State had a population of 17.5 million in 2006 and this grew to 24.0 million in 2016 (Lagos State Government, 2016). Despite its status as a sub-national entity, its demography is as important

as that of a country, for example a Local Government Area (LGA) in the State has more population than a country such as Botswana (Lagos State Government, 2016; UNDP, 2020). Due to this huge population, transmission of RR-TB poses more serious public health concerns particularly within the densely populated settings in the city and the adjoining towns. Given the epidemiological dynamics and risk factors involved in the spread of RR/MDR-TB, there is the need to understand the level of prevalence of RR-TB in such settings in order to improve effective monitoring of patients' treatment as a preventive measure to check the emergence of DR-TB. This study therefore determined the prevalence of RR *Mycobacterium tuberculosis* (MTB) among presumptive pulmonary TB patients in Lagos and its environs and the associated risk factors.

MATERIALS AND METHODS

Study design

A retrospective review of presumptive TB register of patients screened for MTB and RR-TB using Xpert MTB/RIF assay at the Center for Tuberculosis Research Laboratory, NIMR was conducted between January, 2018 and August, 2019. The study population was presumptive TB patients (patients with clinical signs and symptoms suggestive of TB) who reported at the DOTS centers during the period of study.

Laboratory investigation and data collection

A single sputum sample per patient for age greater than six years and a gastric aspirate sample in case of children less than this age group were used in the study for the diagnosis of all presumptive TB patients using Xpert MTB/RIF assay. Samples were collected a wide mouth, dry and sterile container, given to patients. Samples were processed by GeneXpert MTB/ RIF assay. Sample processing including dilution, decontamination and GeneXpert MTB/RIF assay were carried out as earlier reported (Adejumo *et al*., 2018) following the manufacturer's instructions. Patients' with incomplete data set e.g., age, gender, Xpert MTB/RIF results, HIV status, sample type, and location were excluded from the study.

Data analysis

Statistical Package for Social Sciences (SPSS) IBM version 23 was used for data analysis. Measured variables were presented using descriptive statistics such as percentages, mean and standard deviation.

Categorical variables were compared using chi-squared test. *Mycobacterium tuberculosis* (MTB) detection and rifampicin resistance were the outcome variables. Adjusted odds ratio of associated factors of the outcome variable (age, gender, HIV status) were determined. Confidence interval was estimated at 95% level of significance and p <0.05 was considered significant for all statistical tests.

Ethical Consideration

Ethical approval for the study was obtained from the Institutional Review Board (IRB) of the Nigerian Institute of Medical Research (IRB/19/053: 09/10/2019). Confidentiality of information was maintained by de-identifying data retrieved from the registers.

RESULTS

Overall, 1723 presumptive TB patients submitted samples for TB diagnosis, however 1453 (84.3%) had complete data and were included in the study. Out of 1453 patients, 707 (48.6%) were males while 746 (51.3%) were females.

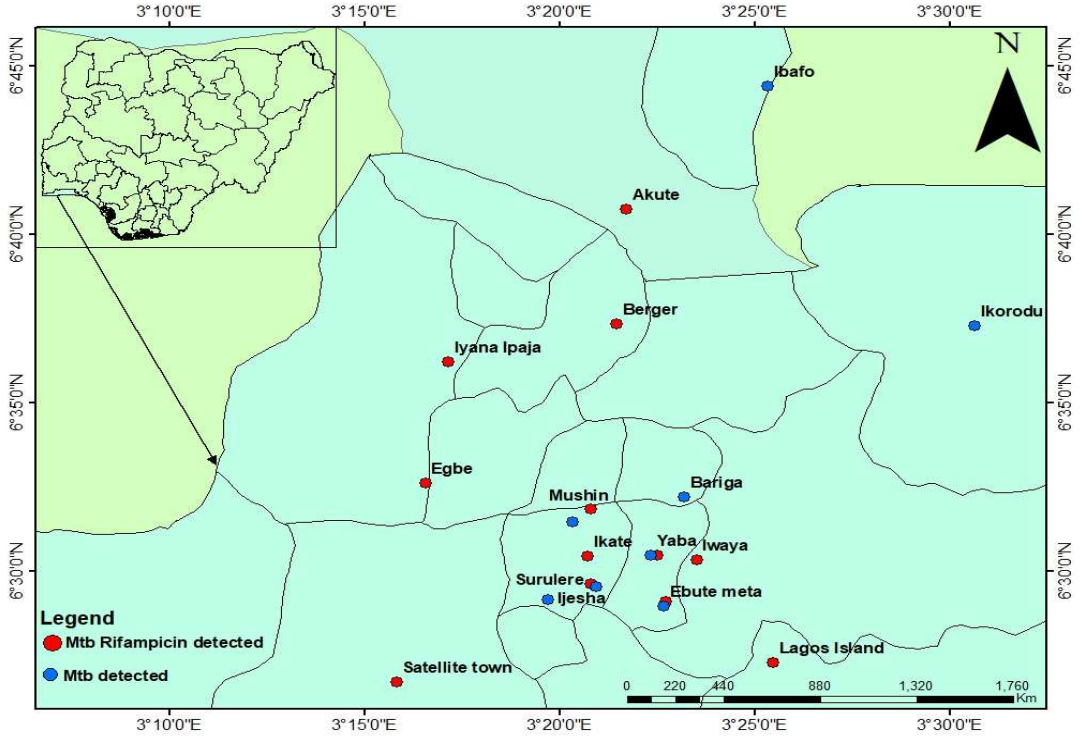

Figure 1. Map of Nigeria showing Lagos State and the affected areas in Lagos State and its environs (Geographical Information System, GIS).

The overall prevalence of TB in the study area was 15.3% with 222 of the 1453 patients being positive for *M. tuberculosis*. Out of the 222 MTB-positive patients, 136 (61.3%) were male while 86 (38.7%) were females. There was however significant relationship between TB and gender (Score test: $\chi^2(1) = 16.752$, $p < 0.001$). The odds of detecting TB were 0.55 times lower for gender (OR 0.547, CI 0.408– 0.733, p < 0.001). The ages of the adult population infected ranged between 16 to 85 years with a prevalence of 99.1%. There was however no significant relationship between TB and age (Score test: $\chi 2$ (1) = 2.981, p > 0.05). The odds of detecting TB were 0.99 times lower for age as a predictor variable (OR 0.992, CI 0.983 – 1.001, p > 0.05). Importantly, the HIV prevalence of the study population was 29.9% while the prevalence of HIV among the study population infected with MTB was 19.4%.

There was significant association between HIV status and TB (Score test: χ^2 (2) = 34.042, p < 0.001). The odds of detecting TB were 1.33 times higher for HIV status as a predictor variable (OR 1.329, CI 0.929 – 1.902, p < 0.01). A prevalence of 13/222 (5.9%) was recorded for RR-TB patients was recorded among the earlier confirmed TB cases (Table 1). Out of the 13 cases, 4 (30.8%) were females while 9 (69.2%) were males. The ages of the affected population ranged between 22 to 75 years (34.85±15.01) while one person (male) was HIV positive.

Table 1. Prevalence of TB and Rifampicin Resistance among the Sample Population

X-pert MTB/RIF Assay	Number (n)	Percentage (%)	LR Test	OR for TB Detected	95% CI for OR
MTB DETECTED (Gender)					
Male	136	61.3	$\chi^2(1)$ =16.752, p < 0.001	0.547	0.408 – 0.733
Female	86	38.7			
Total	**222**	**100**			
MTB NOT DETECTED (Gender)					
Male	571	46.4			
Female	659	53.5			
Total	**1231**	**100**			
MTB DETECTED (HIV Status)					
N	130	58.6			
P	43	19.4	$\chi^2(2)$ = 34.042, p < 0.001	1.329	0.929 – 1.902
U	49	22.1			
Total	**222**	**100**			
MTB NOT DETECTED (HIV Status)					
N	559	45.4			
P	392	31.8			
U	280	22.7			
Total	**1231**	**100**			
MTB DETECTED (Age Group)					
0-15	2	0.9			
16-85	220	99.1	$\chi2(1) = 2.981$ p > 0.05	0.992	0.983 – 1.001
Total	**222**	**100**			
MTB NOT DETECTED (Age Group)					
0-15	117	9.5			
16-85	1112	90.3			
Total	1229	99.8			
MTB DETECTED (Rifampicin Resistance)					
Positive	13	5.9			
Indeterminate	2	0.01			
Negative	207	93.2			
Total	**222**	**100**			

Key: N- Negative, P-Positive, U- Unknown

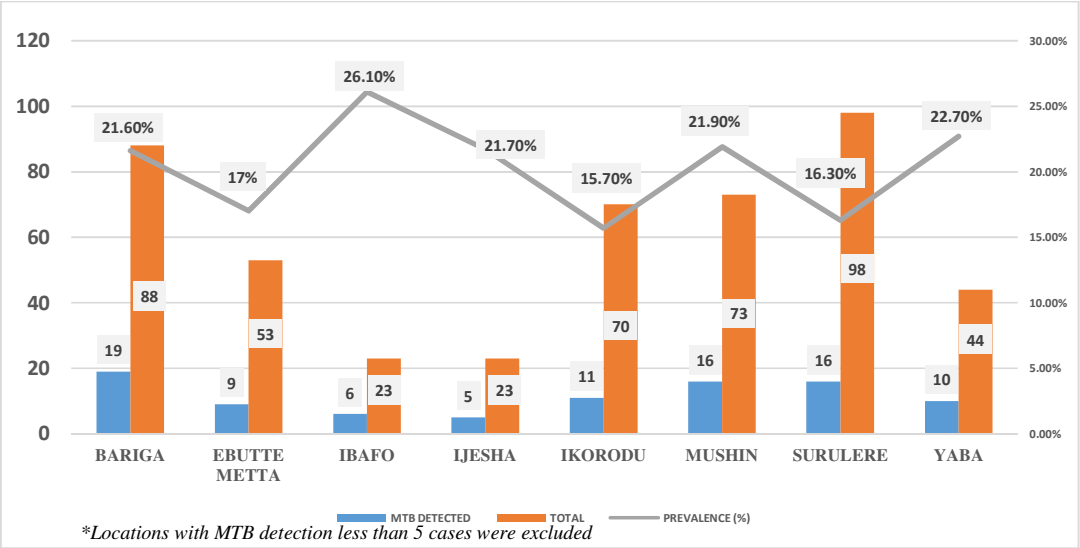

Figure 2. Prevalence of MTB per Location/Residence of Sample Population

MTB infection was detected in 79 locations out of which 8 locations had a high prevalence (15.7% - 26.1%) of the disease. The prevalence of MTB was highest within the sample population who reside in Ibafo (26.1%) followed by residents of Yaba (22.7%) (Figure 2).

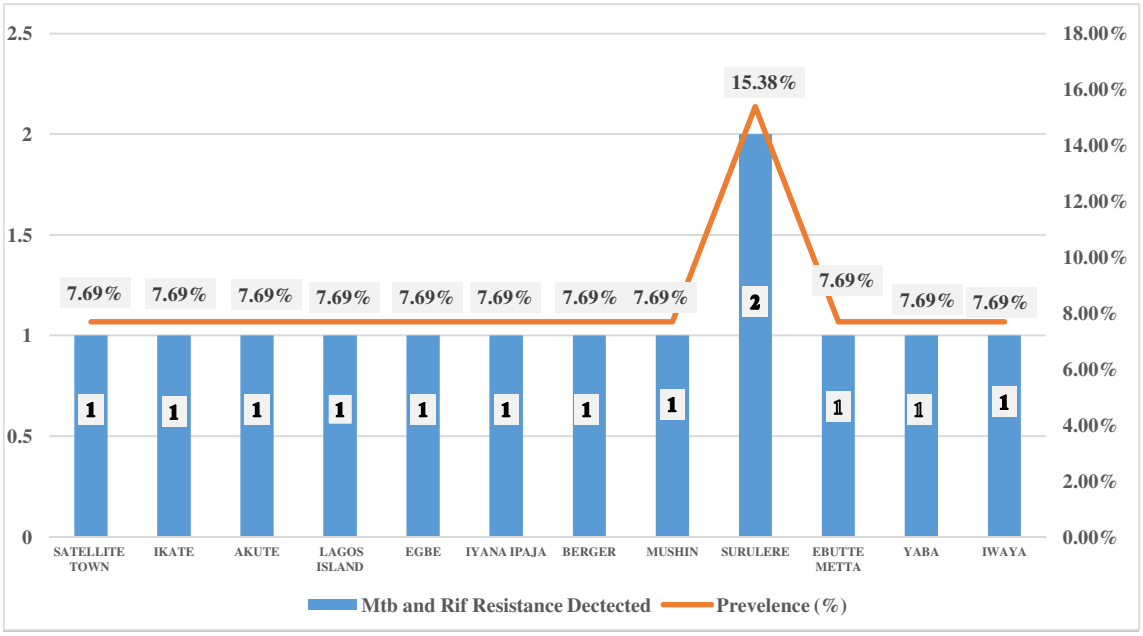

Figure 3. Prevalence of MTB and Rif Resistance per Location/Residence of Sample Population

However, RR-TB was detected in 12 locations and the prevalence of RR-TB was highest within the sample population who reside Surulere, recording the highest prevalence at 15.38%. (Figure 3).

DISCUSSION

Drug-resistant TB continues to pose a serious public health challenge not only in Nigeria but globally despite various TB control programmes and available anti-tuberculosis drugs. Hence there is a need for rapid laboratory TB diagnosis to enable early commencement of treatment so as to limit transmission (Atashi *et al.,* 2017). This study was designed to determine the prevalence of *M. tuberculosis* and rifampicin resistance in new cases of TB in a TB reference laboratory in Lagos. Findings from this study reveal that 15.3% of the patients were positive for TB. This was higher than the 15.1% prevalence reported in a study conducted in Ethiopia (Arega *et al* 2019) but lower than those reported in previous studies conducted in Southern and Northern (Egbe *et al.,* 2016; Adejumo *et al*., 2018; Ikuabe and Ebuenyi, 2018). The variations in prevalence may be attributed to differences such as study design, sample size as well as methodology adopted. In this study, the highest TB prevalence was in Ibafo (26.1%) (Figure 2). Ibafo is a major surburb in Ogun State, South-Western Nigeria. Many people who work in Lagos live in the communities in the outskirts of the city. This is of concern because of ease of transmission through daily commuting in confined mass transit vehicles. Also, DR could be easily acquired in such settings given the overcrowdiness and congested living structure that could facilitate transmission of the disease from infected to healthy people resulting from delayed treatment of those who have developed DR. In this study, more males (61.3%) were infected than females (38.7%). Similar trend has also been reported in previous studies in Nigeria and other African countries (Nyamogoba *et al.,* 2012; Egbe *et al.* 2016; Fadeyi *et al* 2017; Arega *et al* 2019).

The prevalence of HIV and TB co-infection in this study was 19.4%. Our result is higher than previous reports of 12.0% in Southern Nigeria (Onipede *et al* 1999), 10.0% in Northern Nigeria (Iliyasu and Babashami, 2000), 11.6% in Jamaica (Akpaka *et al.* 2006) and 11.4 % in Ethiopia (Tadesse and Tadesse, 2013). These strong relationship between HIV and TB infection has since been established from previous reports. The treatment of both diseases using drug cocktails is typically fraught with challenges that can lead to treatment failure and thus development of drug resistance. The breakdown of immune system and other gastrointestinal opportunistic infections from malabsorption, may also lead to reduced absorption of anti-TB medications, which may further contribute to drug resistance, especially rifampicin (Audu *et al.,* 2017).

Resistance to rifampicin is usually a marker for MDR-TB, hence almost 90% of rifampicin-resistant strains are also resistant to isoniazid (Atashi *et al.,* 2017). In this study, the overall prevalence of RR-TB was 5.9% among the TB confirmed cases. This is higher than the 4.2% reported by Fadeyi *et al.* (2017) in a study conducted in North-Western Nigeria and 2.2% reported by Idigbe *et al*. (1998). The higher prevalence in this study may be due to study setting. This is because the Center for Tuberculosis Research is a National TB reference laboratory where most presumptive DR-TB patients are referred from other hospitals in Lagos State for Xpert MTB/RIF test. In this study, the prevalence of RR-TB was higher in males (69.2%) than females (30.8%). This is similar to previous reports in Nigeria and South Africa (Coovadia *et al.*, 2013; Adejumo *et al.*, 2018). This may be attributed to lifestyle, social habits as well as level of exposure to infection. Surulere had the highest prevalence of RR-TB at 15.38% (2/13) (Figure 3). Its dense population which was estimated at 64,554/ Sq Km in 2016 (Lagos State Government, 2016) is higher than that of Lagos State, Nigeria and Sub-Saharan Africa. This could impact negatively on the person-to-person air exchange necessary for transmission due to crowding and limited ventilation hence increasing rate of transmission (Barun-Mathema *et al*., 2017).

CONCLUSION

Drug resistance is a major cause of public health concern apparently fuelling the TB burden in Nigeria. Drug resistance could be easily acquired due to living in an environment with high prevalence of drug resistance disease. The findings in this study emphasized the importance of drug susceptibility testing in TB management. Consequently, proactive measures are therefore urgently needed to address the issues of prompt diagnosis and early commencement of treatment to reduce the spread of DR-TB in the community. In conclusion, we therefore advocate for the institution of routine TB surveillance through the use of rapid diagnostic tools such as Gene Xpert for early detection and effective treatment management. Since prevalence of RR-TB indicative of a major public health challenge, we advocate for improved and prompt diagnosis, as well as effective monitoring of patients' treatment to prevent further emergence and spread of drug resistant tuberculosis in the country.

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