



Retrospective Analysis of Rifampicin-resistance Pattern of *Mycobacterium tuberculosis* among Presumptive Tuberculosis Patients in Secondary Referral Hospital Offa, Nigeria

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Authors' contributions

This work was carried out in collaboration among all authors. Authors PDA and OSA designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors MU and IAA managed the analyses of the study. Authors MGO, DKS and OOA managed the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Introduction: Early detection of drug resistant tuberculosis with more sensitive and accessible method together with appropriate control measures among other factors are keys to interruption of transmission and mortality rate.

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Study Design: This study conducted a retrospective review of presumptive TB register of patients screened for *Mycobacterium tuberculosis* (MTB) and rifampicin resistant tuberculosis (RR-TB) using Xpert MTB/RIF assay at General Hospital Offa in Kwara State Nigeria between January to December 2018 in order to access the on-going efforts such as quarterly surveillance, improved infrastructural facilities and intense media advocacy on Tuberculosis by Kwara state government on prevention of Tuberculosis in Kwara State.

Results: Overall, 63 (10.6%) of the sputum samples collected were positive for *Mycobacterium tuberculosis* while 4 (6.3%) showed rifampicin resistance out 63 (10%) of patients positive for *Mycobacterium tuberculosis*. There is high recovery rate of *Mycobacterium tuberculosis* among women than men as this study recovers 52.4% and 47.6% in male and female respectively. Adults appear to be more vulnerable to TB and the vulnerability tends to increase with age.

Conclusion: This study documents low frequency of rifampicin resistance tuberculosis particularly within female population using highly sensitive and specific Xpert MTB/RIF in Kwara State.

Keywords: Retrospective; rifampicin resistance; *Mycobacterium tuberculosis*; Offa.

1. INTRODUCTION

Multidrug resistant TB (MDR-TB), defined by *Mycobacterium tuberculosis*, resistant to isoniazid and rifampicin (RIF), the two key first-line anti-TB drugs in short course chemotherapy [1,2]. MDR-TB is an increasing global health problem [3-8]. While MDR-TB develops as a consequence of poor adherence to anti-TB treatment [1,2], Person-to-Person transmission can also occur particularly among health workers (22). The survey based on modeling predicts MDR-TB prevalence in Nigeria to vary from 4.3% (3.2-5.4%) for new cases, 25% (19-31%) for retreated cases [9]. The universal control of tuberculosis is hindered by slow, insensitive diagnostic methods, especially for the identification of drug-resistant forms. Early diagnosis is crucial to lower the mortality rate and halt transmission, but the complexity and infrastructure needs of sensitive methods hinder their accessibility and consequence [1,4]. The common methods of drug resistance testing involve isolation of *M. tuberculosis* on liquid or solid culture medium and with turnaround time of about 8 weeks [1]. Culture methods are time-consuming and expensive consequently reducing both satisfaction and likelihood of prompt treatment for patients [10,11]. Recently, different modern commercial tests based on the genetic sequence have been described for identification of MDR-TB. Example of such is used in the current research for identification of both TB and drug resistant TB [12]. It definitely detects genetic mutations that confer resistance to both isoniazid and rifampicin in patients' samples. It produces an option to traditional drug sensitivity testing through culture because it is cheap and effective [13,10]. Globally, several locations have

used this test successfully with high sensitivity rates for RIF (>95.5%) and isoniazid (>81.8%) resistance [9-16] and 100% specificity [9-16]. Recently, this new nucleic acid amplification diagnostic technologies has attracted much attentions due to their sensitive, specific and swift nature – the Genexpert MTB/RIF. The Xpert assay utilizes, heminested real-time polymerase chain reaction (PCR) to amplify specific rpo B gene of *M. tuberculosis* [17,18]. To detect rifampicin resistance, molecular beacons is used to probe the rifampicin resistance – determining region of rpoB gene is [19]. Therefore, identification of resistance to rifampicin can be accomplished within two hours. The aim of this study is to determine the prevalence of rifampicin resistant TB in our locale using the automated Gene Xpert test system.

2. METHODS

The study was performed in TB Referral Hospital (General Hospital, Offa), located at Southern region of Kwara State. The state covers an area of 36,825 square kilometres. It has a population of 2,365,353 (based on the 2006 census figures). It accounts for 1.69% of Nigeria's total population [16]. The hospital serves the state, neighboring states and also serves as a referral hospital in the entire Kwara South. All sputum specimens that were submitted to the TB referral hospital laboratory from January to December 2018 of patients who were 4 years and above were included in the study.

XPERT procedure (MTB/RIF assay): Sample reagent was added in a 2:1 ratio to untreated sputum and in a 3:1 ratio to decontaminate sputum pellets. The additional sample reagent in

pellets was necessary to meet the volume requirements for the assay sample. The closed sputum container was manually agitated twice during a 15-minute period at room temperature before 2 mL of the inactivated material was transferred to the test cartridge (equivalent to 0.7 mL of untreated sputum or 0.5 mL of decontaminated pellets). Cartridges were inserted into the test platform (Cepheid, Sunnyvale, CA) is an integrated diagnostic device that performs sample processing and heminested real-time polymerase chain reaction (PCR) analysis in a single hands-free step for the diagnosis of tuberculosis and rapid detection of RIF resistance in specimens. The electronic results were sent directly from the MTB/RIF test system to the central database and read after 90 minutes.

2.1 Statistical Analysis

This was by sample percentages using Microsoft Excel 2010 for Windows (Redmond, CA, USA).

3. RESULTS

This study included n=597 sputum specimens, all of which were sent to TB Referral Hospital (General Hospital, Offa) between January to December 2018. Altogether, 63 (10.6%) of the sputum samples collected were positive for *M. tuberculosis* (Table 1); with 4 (6.3%) showed rifampicin resistance (Table 2). TB appeared to

be more prevalent in November with the highest prevalence recorded in December (Fig. 1). In addition, the prevalence of TB and MDR-TB appeared to be increased in > 16 years and above (Fig. 2).

4. DISCUSSION

MDR-TB is TB with bacilli resistant to at least isoniazid and rifampicin, the main anti-tuberculosis drugs. It has become an important concern for TB control in many countries, especially in low-income countries where the burden of other competing diseases like malaria, enteric fever, meningitis and other diseases is high [20,21]. With the advent of the Xpert MTB/RIF testing it is now readily possible to rapidly determine TB bacilli susceptibility to common anti-TB drugs [20,22]. Drug resistant TB develops from the inadequate treatment of active pulmonary TB. There are multiple reasons for inadequate therapy; poor prescribing practices with insufficient treatment duration, far distance between health facilities and patient's area of residence as well as poor drug selection are well-recognized contributory factors [1,20]. Similarly, Fundamental problems such as; inadequate public health resources, inadequate drug supplies, patient education, adverse events or socio-economic status also contribute. Report has also shown significant number of people acquire drug resistant strain due to location of their residence in high prevalence of drug resistance area [20].

Table 1. Frequency of *M. tuberculosis* (MTB) detection by GeneXpert

| | Frequency | Percentage |
|--------------|-----------|------------|
| MTB Positive | 63 | 10.6 |
| MTB Negative | 534 | 89.4 |
| Total | 597 | 100 |

Table 2. Frequency of *M. tuberculosis* (MTB) resistance to Rifampicin

| | Frequency | Percentage |
|-----------------------|-----------|------------|
| Rifampicin sensitive | 59 | 93.7 |
| Rifampicin resistance | 4 | 6.3 |
| Total | 63 | 100 |

Table 3. Frequency of MTB positive with respect to gender

| | Frequency | Percentage |
|--------|-----------|------------|
| Male | 33 | 52.4 |
| Female | 30 | 47.6 |
| Total | 63 | 100 |

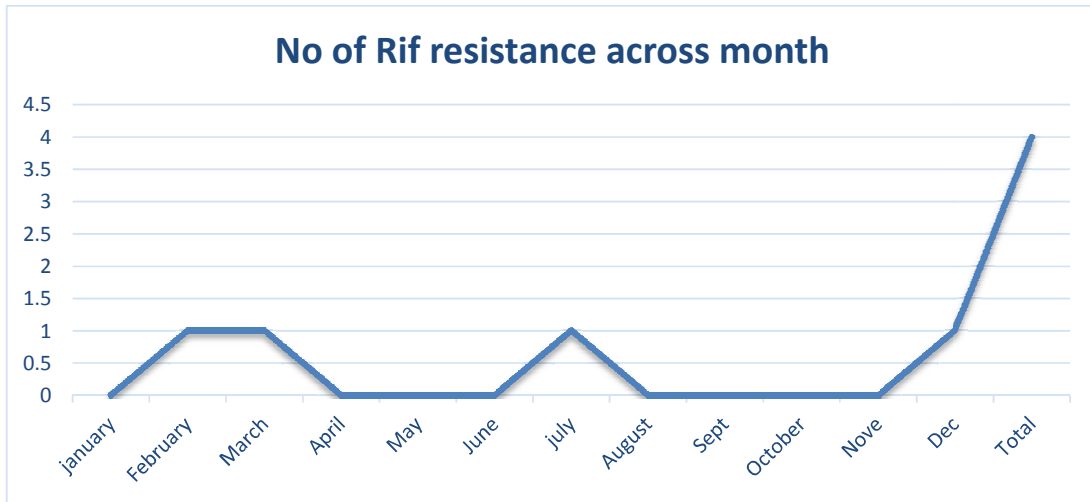


Fig 1. Prevalence of Rifampicin resistance tuberculosis across months

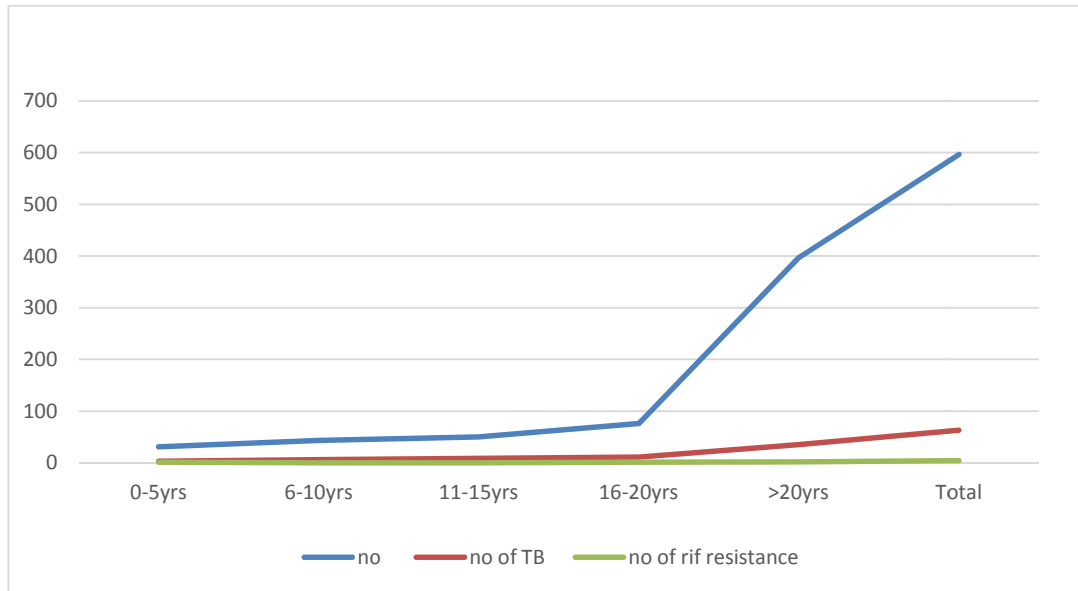


Fig. 2. Prevalence of pulmonary tuberculosis and Rifampicin resistance tuberculosis across different age groups

Using the Xpert MTB/RIF assay system, this study obtained frequency of 6.3% rifampicin mono resistance. This is consistency with the study carried out by Otu A et al., [23] where they attributed low frequency of rifampicin mono-resistance to the new history of use of rifampicin in African countries [24]. Furthermore, our prevalence frequency of 10.6% MTB positive and 6.3% for rifampicin resistance is a good indicator that TB prevalence in Kwara South is very low when compared with studies conducted in Lagos by Adejumo et al., [24] which recorded 37.7% for MTB positive with 23.4% for rifampicin

resistance and also lower than 43.3% MTB positive and 17.6% for rifampicin resistance recorded by Kuyinu et al. [25] carried out in Lagos state.

Similarly, our result of 10.6% and 6.3% for MTB positive and rifampicin resistant had further proved the low prevalence of MTB and rifampicin resistance in Kwara South when compared with study carried out in Nasarawa(North Central) by Egbe et al. [26] which reported a prevalence rate of 57.1% for MTB positive while 6.1% was observed for rifampicin resistance and the study

conducted by Ikuabe et al., [27] in Yenogoa (South East) where the prevalence rate stood at 22.9% and 14.7% for both MTB positive rifampicin resistance respectively.

The risk of transmission of TB does not appear to be more in spring and summer in this study as this study observed a progressive increase prevalence of TB and MDR-TB between Novembers to December. This is consistent with work done in Wuhan, China that seasonal variation occurs in the prevalence of pulmonary tuberculosis. This may be due to overcrowding of people in different households in Kwara State during winter, which could lead to an increase in transmission of TB, an observation reported by Xiaobing et al. [28] in a study done in China.

Similarly, age appears to be another important risk factor for TB and Drug Resistance TB as this study examined a progressive increase prevalence of TB for age 16 years and above. This is in agreement with the WHO (2019) report that TB accounts for 89% in >15 years age group.

There is need to strengthen global management of tuberculosis by stepping up efforts in the development of rapid diagnostic test for MDR-TB for the purpose of rapid detection and treatment of MDR-TB. Limitation of this study is that it was carried out only in TB Referral Hospital in General Hospital, Offa and its catchment area. A larger area survey both at national and state level will provide a better estimate of MDR-TB in Nigeria.

There is need to increase the laboratory capacity for rapid detection of MDR-TB in many more zones of the Kwara State and in Nigeria for effective wider coverage. There is also need for this kind of research to be replicated in the remaining two zones (Kwara Central and Kwara North) of the state in order to have a complete and reliable data on MTB in Kwara State.

5. CONCLUSION

There is hope that *Mycobacterium tuberculosis* can actually be eradicated in Kwara south or reduced to barest minimum. The low frequency of rifampicin resistance also proved that, provided the present cases strictly adhere to their treatment regimen, there is likelihood of achieving low incident of *Mycobacterium tuberculosis* in Kwara South in the nearest future.

CONSENT

As per international standard or university standard, patient's written consent has been collected and preserved by the author(s).

ETHICAL APPROVAL

This study was approved by the Research and Ethics Committee of the State Ministry of Health.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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