

## ORIGINAL ARTICLE

# Trends and Prevalence of Rifampicin-Resistant Tuberculosis at Ndola Teaching Hospital, Zambia (2020–2022)

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## ABSTRACT

**Background:** Rifampicin resistance tuberculosis (TB) is a major public health problem in Zambia. It is associated with Multidrug-resistant TB, caused by *Mycobacterium tuberculosis* strains resistant to at least rifampicin, one of the most potent first-line anti-TB drugs. Rifampicin resistance (RR) TB represents a significant threat to TB control efforts, complicating treatment by necessitating longer, more toxic, and costly regimens with poorer outcomes. This poses considerable challenges for both diagnosis and treatment, further straining Zambia's healthcare system. The study aimed to determine the trends and prevalence of rifampicin-resistant Tuberculosis at Ndola Teaching Hospital, Zambia (2020–2022).

**Method:** A laboratory-based, retrospective, cross-sectional study was conducted at Ndola Teaching Hospital, spanning a three-year period from January

2020 to December 2022. The study included all patients with clinically suspected TB whose sputum samples were tested using the GeneXpert MTB/RIF assay. The sample size was calculated using Cochran's formula, based on an estimated RR-TB prevalence of 5.9%, resulting in a minimum required sample size of 2,133 participants. Sputum samples were processed using the GeneXpert MTB/RIF assay, which detects *Mycobacterium tuberculosis* and RR through real-time polymerase chain reaction. Ethical approval was granted by the University of Zambia School of Medicine Undergraduate Research Ethics Committee and the National Health Research Authority. Descriptive statistics were employed, and data analysis was carried out using the Statistical Package for Social Sciences (SPSS) version 22.

**Results:** The study found an overall TB prevalence of 12% (95% CI: 11.15%–12.86%) among 5,590 participants, with RR-TB detected in 2.98% of cases. RR-TB prevalence was highest among males and the 18–34 age group 60% (95% CI: 38.66%–78.12%). RR-TB prevalence increased

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significantly from 1.53% (95% CI: -0.19–3.20) in 2020 to 3.13% (95% CI: 0.84–5.36) in 2021 and 4.6% (95% CI: 1.89–7.05) in 2022, contrasting with the stable trend in overall TB prevalence 11.41% (95% CI: 9.92–12.90) in 2020, 11.96% (95% CI: 10.50–13.43) in 2021, and 12.57% (95% CI: 11.10–14.04) in 2022. HIV/TB co-infection was observed in 36.8% of participants, highlighting the ongoing burden of dual infection.

**Conclusion:** This study highlights the growing burden of RR-TB, despite stable overall TB rates. The increase in RR-TB, particularly among males and young adults, underscores the urgent need for enhanced diagnostic capacity, including widespread use of rapid molecular tests like GeneXpert, and targeted interventions for high-risk groups. Integrated HIV/TB care and improved access to second-line treatments are critical to address the clinical implications of rising RR-TB, which complicates treatment and increases healthcare costs.

## INTRODUCTION

Tuberculosis (TB) remains a significant public health challenge in Zambia, with an estimated 59,000 cases recorded in 2020, translating to an incidence rate of 319 cases per 100,000 population<sup>1</sup>. While Zambia has made progress in reducing TB-related mortality from 21.3 per 100,000 in 2011 to 12.7 per 100,000 in 2019<sup>2</sup>. TB remains a leading cause of death, particularly due to its strong association with HIV co-infection, which affects approximately 59% of new and relapse TB cases<sup>3</sup>. Despite progress, significant challenges persist, including limited access to quality laboratory services, low public awareness, stigma, and the growing threat of drug-resistant TB.

A growing concern in TB management is the emergence of drug-resistant TB, particularly rifampicin-resistant TB (RR-TB), which is a key marker of multidrug-resistant TB (MDR-TB)<sup>3</sup>. RR-TB complicates treatment by necessitating longer, more toxic, and costly regimens with poorer

treatment outcomes<sup>3</sup>. Despite Zambia's efforts to scale up rapid diagnostic tools such as the Xpert MTB/RIF assay and expand access to second-line TB treatment, significant challenges remain, including delays in diagnosis, suboptimal treatment adherence, and healthcare system inefficiencies<sup>4</sup>. Additionally, the high burden of HIV further exacerbates the challenge, as immunocompromised individuals are at a 19- to 20-fold higher risk of developing active TB compared to those who are HIV-negative<sup>5</sup>.

While previous studies have documented the burden of RR-TB in Zambia at the national level, there is limited facility-based data on its trends, demographic patterns, and risk factors, particularly in the Copperbelt region. Existing estimates suggest that RR-TB and MDR-TB rates are significantly higher among previously treated TB patients, with national figures indicating that approximately 1.1% of new TB cases and 18% of retreatment cases are MDR-TB<sup>6,7</sup>. However, provincial and facility-level variations remain largely uncharacterized. A study by Masenga et al. (2017) reported an RR-TB prevalence of 5.9%<sup>3</sup>, whereas the World Health Organization (WHO) reported a lower national prevalence of 2.8%<sup>7</sup>, highlighting regional disparities that warrant further investigation. Like many other countries, Zambia was significantly impacted by the COVID-19 pandemic, which further constrained TB control efforts. The disruptions to healthcare services, delays in diagnosis, and interruptions in treatment regimens contributed to the rise in drug-resistant TB cases<sup>8</sup>. In the post-COVID-19 era, understanding the impact of these challenges is essential for developing effective strategies to mitigate the long-term consequences on TB control.

This study is particularly significant as no previous investigation has specifically examined RR-TB in Ndola, a major city in Zambia's Copperbelt province. The study aimed to address this lack of information by assessing the prevalence and trends of RR-TB at Ndola Teaching Hospital (NTH), a key referral

centre in the region. Covering a three-year period (2020–2022), the study investigates RR-TB prevalence while analysing demographic and clinical risk factors such as age, gender, and HIV status. By adopting a facility-based approach, this research provides targeted insights that can strengthen TB control efforts, enhance diagnostic capacity, and inform policy interventions to address the growing challenge of RR-TB in Zambia

## MATERIALS AND METHODS

### Study design, site and population

This was a laboratory-based, retrospective, cross-sectional study conducted at Ndola Teaching Hospital (NTH) from January 2020 to December 2022. The study included all patients with clinically suspected TB whose sputum samples were referred to the NTH laboratory for GeneXpert MTB/RIF testing. NTH is a referral hospital and multi-drug resistance centre located in the Copperbelt region of Zambia. The study population was limited to patients whose samples were tested using GeneXpert, as this was the primary diagnostic tool for TB and rifampicin resistance at NTH during the study period.

### Inclusion and Exclusion criteria

This study included all patients with clinically suspected TB whose sputum samples were analysed using the GeneXpert MTB/RIF assay at NTH between January 2020 and December 2022. Patients of all age groups and genders were eligible, provided their records contained complete demographic and laboratory test results, including TB results, rifampicin resistance result, and HIV status (positive, negative, or unknown)<sup>3</sup>. However, patients were excluded if their samples were not tested using GeneXpert MTB/RIF, if their records had incomplete or missing data on key study variables, or if they had contaminated or invalid test results that could not be used for analysis.

### Sample size

The sample size for this study was calculated using Cochran's formula, for cross-sectional studies, based on the total number of TB presumptive cases tested at NTH with the GeneXpert MTB/RIF assay from 2020 to 2022. With a reported RR-TB prevalence of 5.9% in prior Zambian studies Masenga et al., (2017)<sup>3</sup>, the sample size was determined to ensure sufficient statistical power to detect RR-TB prevalence within this range.

$$N = \frac{Z^2 P(1 - P)}{d^2}$$

where:

- N = required sample size
- Z = Z-score for a 95% confidence level (1.96)
- P = estimated prevalence of rifampicin-resistant TB (assumed 5.9% or 0.059 based on prior reports)
- d = margin of error (0.01 or 1%)

$$N = \frac{(1.96)^2(0.059)(0.941)}{0.01^2}$$

$$N = 2,132.8$$

The minimum required sample size for detecting rifampicin resistance was 2,133 participants. However, to increase the study's statistical power and account for potential missing or incomplete data, all available 5,590 cases were included. This larger sample size enhances the reliability of the prevalence estimates and allows for more robust subgroup analyses by age, gender, and HIV status.

### Sample processing

Sputum samples collected from patients with clinically suspected TB were processed at the NTH Laboratory using the GeneXpert MTB/RIF assay.

Upon receipt, samples were logged into the Laboratory Information System (Disa) and assigned unique identification numbers to ensure proper tracking and prevent duplication. Sample quality was assessed for adequate volume, consistency, and contamination before testing.

The GeneXpert MTB/RIF assay, which is a nucleic acid amplification test, was conducted utilizing the GeneXpert Dx 4.7b Software System along with single-use cartridges (Cepheid, Sunnyvale, California, USA)<sup>9</sup>.

Each sample was processed as per the manufacturer's instructions of the GeneXpert MTB/RIF by trained Laboratory technologist and Scientists. A buffer reagent was added to the sputum sample in a 2:1 ratio, ensuring proper liquefaction and inactivation of MTB. The mixture was then vortexed for 15 minutes to ensure homogeneity before being transferred into the GeneXpert cartridge<sup>10</sup>. The cartridges were loaded into the GeneXpert IV system, which performed automated real-time polymerase chain reaction (PCR) to detect MTB and RR.

Results were automatically generated within 90 minutes and reviewed by laboratory personnel. Positive results were classified as MTB detected, MTB not detected, or MTB detected with rifampicin resistance. Internal quality control (IQC) measures, including the use of negative and positive controls, were conducted periodically to ensure assay reliability. Invalid or indeterminate results were retested, and if issues persisted, patients were requested to provide a new sputum sample for repeat analysis.

The Xpert MTB/RIF assay includes built-in quality controls to ensure accurate specimen processing, successful PCR amplification, and cartridge functionality. Each cartridge is equipped with a Sample Processing Control (SPC), which contains non-infectious spores in a dry spore cake. The SPC verifies effective MTB lysis, assesses the adequacy of specimen processing, and detects potential inhibition of the real-time PCR assay<sup>11</sup>. In negative

samples, the SPC must be positive, while in positive samples, it may be either positive or negative. A Probe Check Control is conducted prior to PCR initiation to evaluate fluorescence signals, confirming proper bead rehydration, reaction-tube filling, probe integrity, and dye stability. The IQC and inbuilt controls collectively ensure the reliability and accuracy of the assay.

### **Data collection**

Data was collected using an Excel spreadsheet from the Laboratory Information System and TB registers at NTH Laboratory. The collected data included the year of diagnosis, age, gender, HIV status, TB results, and rifampicin outcomes. The data was then transferred to Statistical Package for Social Sciences (SPSS) version 22 for statistical analysis. Cases that did not meet the inclusion criteria such as incomplete data and missing information were excluded from the study.

### **Data analysis**

After data collection, records were de-identified and subsequently transferred to SPSS version 22 for statistical analysis. Descriptive statistics were initially applied to calculate frequencies and percentages for categorical variables, including gender and HIV status. For inferential analysis, a chi-square test was conducted to determine the statistical significance of rifampicin resistance among confirmed MTB cases. A 95% confidence interval was used, and statistical significance was considered at a p-value of less than 0.05.

### **Bias Minimization**

To ensure the validity and reliability of the findings, several measures were implemented to minimize potential biases in this study. Selection bias was reduced by including all patients with clinically suspected TB who had their samples analysed using the GeneXpert MTB/RIF assay at NTH between January 2020 and December 2022, ensuring a comprehensive and representative sample. Information bias was minimized by using standardized data collection procedures from the

Laboratory Information System and TB registers, ensuring consistency in recording patient demographics, TB status, RR, and HIV status. Observer bias was addressed by anonymizing patient records during data extraction and analysis, preventing investigator influence on data interpretation. Confounding factors such as age, gender, and HIV status were accounted for through stratified analysis, enabling a clearer assessment of their impact on TB and rifampicin resistance trends.

Unknown HIV status was included as a separate category in descriptive analyses and statistical tests comparing HIV-positive and HIV-negative groups. This category also encompassed participants with missing HIV status information, who were initially classified as having an unknown HIV status. Additionally, to prevent data duplication, patient records were carefully cross-checked using unique identifiers before analysis, ensuring that each case was counted only once. These measures strengthened the study's internal validity and enhanced the generalizability of the findings.

**Ethical approval**

Ethical approval for this study was obtained from the Mulungushi University School of Medicine Research Committee (IRB: 00012281, FWA: 0002888) under reference number SMHS-MU1-2024-04, as well as the National Health Research Authority on 12th January 2024 and 7th February 2024. Additionally, permission to conduct the study was granted by the Ndola Teaching Hospital Management. To maintain patient confidentiality and data security, all patient records were anonymized by assigning unique study identification numbers before data extraction and analysis. Personal identifiers such as names, national identification numbers, and contact details were removed from the dataset. Data was stored in a password-protected electronic database accessible only to authorized research personnel. Hard-copy records, if used, were securely stored in a locked cabinet within the hospital laboratory, with access restricted to designated team members. All research

activities adhered to the principles of the Declaration of Helsinki and relevant national research ethics guidelines to ensure that participant privacy was safeguarded.

**RESULTS**

**Demographics of participants**

Among the 5,590 patients analysed, the age distribution showed that 1,287 (23.0%) were in the 35–44 years age group, 1,258 (22.5%) were in the 25–34 years group, and 1,018 (18.2%) were in the 45–54 years group (Table 1). Of the total sample, 3,241 (58%) were male and 2,349 (42%) were female. Regarding HIV status, 2,507 (48.8%) were HIV-negative, 1,055 (36.8%) were HIV-positive, and 1,029 (18.4%) had an unknown HIV status.

**Table 1. Characteristic of Participants**

Characteristics	Total (N = 5,590) n (%)	
	Age groups (years)	< 17
18 – 24		485 (8.7)
25 – 34		1258 (22.5)
35 – 44		1287 (23.0)
45 – 54		1018 (18.2)
55 – 64		674 (12.1)
> 65		798 (14.3)
Gender	Female	2349 (42.0)
	Male	3241 (58.0)
HIV Status	Positive	2055 (36.8)
	Negative	2507 (48.8)
	Unknown	1029 (18.4)

**Prevalence of TB Disease**

Out of 5,590 samples, *Mycobacterium tuberculosis* was detected in 671 cases, representing a positivity rate of 12% (95% CI: 11.15%–12.86%). The highest prevalence was observed in the 18–24 age group at 19.38% (95% CI: 15.86%–22.9%), followed by the 25–34 age group at 16.53% (95% CI: 14.48%–18.59%). As shown in Table 2.



**Table 2. Prevalence of TB Disease by gender and age group**

Characteristics		Total N	TB Disease N	Prevalence (95% CI)	95% CI Lower (%)	95% CI Upper (%)
	<b>Overall</b>	5590	671	12.00	11.15	12.8
<b>Age groups (years)</b>	< 17	70	10	14.29	6.09	22.48
	18 – 24	485	94	19.38	15.86	22.9
	25 – 34	1258	208	16.53	14.48	18.59
	35 – 44	1287	177	13.75	11.87	15.63
	45 – 54	1018	111	10.9	8.99	12.82
	55 – 64	674	33	4.9	3.27	6.53
	> 65	798	38	4.76	3.28	6.24
<b>Gender</b>	Female	671	221	32.94	29.36	36.51
	Male	671	450	67.06	63.49	70.64

**Prevalence of Rifampicin resistance among presumptive tuberculosis patients**

Out of 5,590 individuals tested, 671 (12%; 95% CI: 11.15%–12.86%) were identified as presumptive tuberculosis cases. Among these 671 TB participants, rifampicin-resistant *Mycobacterium*

*tuberculosis* was detected in 20 cases, representing a prevalence of 2.98% (95% CI: 1.79%–4.32%), as shown in Table 3. Males had a higher prevalence of 65% (95% CI: 45%–85%) compared to females at 35% (95% CI: 15%–55%), with a p-value of 0.058.

**Table 3. Prevalence of Rifampicin resistance among presumptive tuberculosis patients**

Characteristics		Total N	TB Disease N	Prevalence (95% CI)	95% CI Lower (%)	95% CI Upper (%)
	<b>Overall</b>	20	671	2.98	1.79	4.32
<b>Age groups (years)</b>	< 17	0	20	0	0	0
	18 – 34	12	20	60	38.66	78.12
	35 – 54	6	20	30	10	50
	55 – 64	1	20	5	0	15
	> 65	1	20	5	0	15
<b>Gender</b>	Female	7	20	35	15	55
	Male	13	20	65	45	85

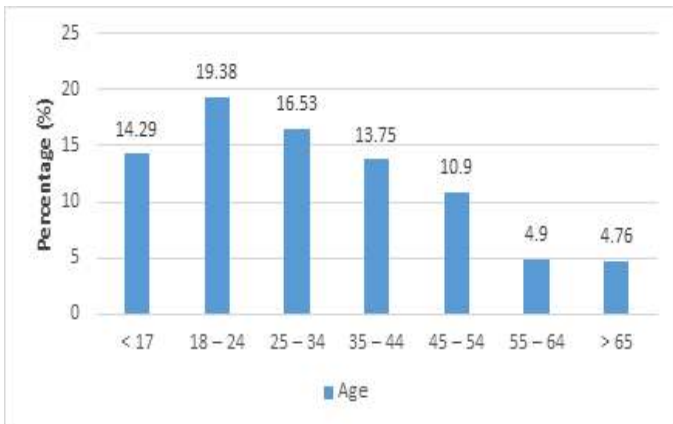
### Tuberculosis positivity trends by year

uring the study period from 2020 to 2022, the overall prevalence of Tuberculosis among tested individuals was 11.41% (95% CI: 9.92–12.90) in 2020, 11.96% (95% CI: 10.50–13.43) in 2021, and 12.57% (95% CI: 11.10–14.04) in 2022. In contrast, the prevalence of rifampicin resistance Tuberculosis among confirmed Tuberculosis cases increased from 1.53% (95% CI: -0.19–3.20) in 2020 to 3.13% (95% CI: 0.84–5.36) in 2021 and 4.6% (95% CI: 1.89–7.05) in 2022. A chi-square test was performed to compare Tuberculosis prevalence across the three years ( $\chi^2 = 0.028$ ,  $p = 0.986$ ) while for rifampicin resistance Tuberculosis was ( $\chi^2 = 4.82$ ,  $p = 0.09$ ) as depicted in Table 4 and figure 3 below.

**Table 4. Tuberculosis prevalence by year trends**

Year	TB Prevalence (%)	95% CI	RR-TB Prevalence (%)	95% CI
2020	11.41	9.92–12.90	1.53	-0.19–3.20
2021	11.96	10.50–13.43	3.13	0.84–5.36
2022	12.57	11.10–14.04	4.60	1.89–7.05

The highest prevalence was observed in the 18–24 age group at 19.38% (95% CI: 15.86%–22.9%), followed by the 25–34 age group at 16.53% (95% CI: 14.48%–18.59%) as depicted in Fig 1.

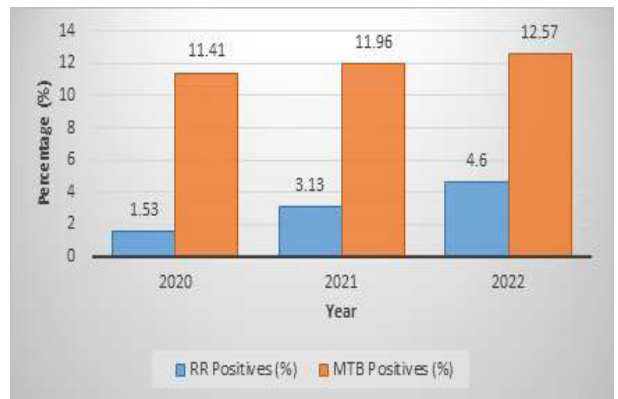


**Figure 1. Tuberculosis prevalence by Age**

The prevalence of tuberculosis by gender revealed a higher rate in males, accounting for 67.06% (95% CI: 63.49%–70.64%), compared to 32.94% (95% CI: 29.36%–36.51%) in females, as shown in Fig. 2



**Figure 2. Tuberculosis prevalence by Gender**



**Figure 3. Tuberculosis prevalence by year trends**

The figure below illustrates the trends in tuberculosis prevalence over the study period from 2020 to 2022. Overall, the prevalence of tuberculosis among tested individuals was 11.41% (95% CI: 9.92–12.90) in 2020, 11.96% (95% CI: 10.50–13.43) in 2021, and 12.57% (95% CI: 11.10–14.04) in 2022. In contrast, the prevalence of rifampicin-resistant tuberculosis among confirmed tuberculosis cases steadily increased, rising from 1.53% (95% CI: -0.19–3.20) in 2020 to 3.13% (95% CI: 0.84–5.36) in 2021, and reaching 4.6% (95% CI: 1.89–7.05) in 2022. As depicted in fig 3 above.

## DISCUSSION

In this study, the prevalence of RR-TB, as detected using the GeneXpert MTB/RIF assay, was 2.98% among 671 TB cases recorded between 2020 and 2022. This finding is consistent with the 3%<sup>12</sup> prevalence reported in the 2021 Ministry of Health National TB and Leprosy Programme report and the 2.8% reported by the WHO<sup>7</sup>. However, it is lower than the 5.9%<sup>3</sup> prevalence observed in a similar study by Masenga et al (2017). Conversely, it exceeds the 1.4%<sup>4</sup> prevalence reported in Zambia's Central Province by Chanda (2024). The observed variations may emanate from differences in sample sizes, geographic locations, and challenges in data capture and record-keeping, as noted in previous studies. Contributing factors include disparities in diagnostic access and healthcare infrastructure, differences in regional HIV/TB co-infection rates, and disruptions to TB services caused by COVID-19, which led to delays in diagnosis and treatment during this study period.

Our study findings indicated that males had a significantly higher prevalence 65% of RR-TB compared to females 35%, consistent with the previous studies as well as results of the TB National Survey in Zambia<sup>3,12-14</sup>. With a p-value of 0.058, we failed to reject the null hypothesis, indicating insufficient evidence to conclude a significant difference in RR-TB prevalence between males and females<sup>3</sup>. However, there was no similar study that had reported a predominance of TB or RR-TB among females. This aligns with global and regional trends where men consistently exhibit higher TB prevalence due to occupational exposures, delayed healthcare-seeking behaviour, and lifestyle factors such as tobacco and alcohol use. Addressing these gender-specific barriers is essential for effective TB control. Implementing targeted outreach programs to improve healthcare access for men, alongside community education campaigns to reduce stigma and promote early diagnosis, could help mitigate these disparities.

Age-specific analysis in our study showed that TB prevalence was highest in the 18–24 age group (19.38%), followed by the 25–34 age group (16.53%). However, the small number of RR-TB (n=20) limited the depth of our analysis, particularly when stratifying results by age groups. For example, some age groups (e.g., <17 years and 35–44 years) had only one or zero RR-TB cases, resulting in uninformative confidence intervals (e.g., 95% CI: 0–0). To address this limitation, we excluded the <17 years age group and combined the 18–24- and 25–34-years categories into a single group (18–34 years). This approach ensured a more meaningful analysis and minimized the risk of overinterpreting age-specific trends. There was limited data on RR-TB as an age-related risk factor. Therefore, our study analysed age-specific factors by referencing previous studies on MDR-TB in Zambia that included RR-TB data. This approach allowed us to contextualize our findings within existing research and identify potential trends in age-related RR-TB susceptibility. Our study found that the highest prevalence of RR-TB was in the 18–34 years age group (60%), which contrasts with findings by Monde et al., (2021) who reported the highest rates in the 30–49 years age group<sup>15</sup>, and Chanda (2024), who identified peak prevalence in the 26–45 years age group<sup>4</sup>. These variations may be influenced by differences in sample size, study design, population demographics, underlying risk factors, and the healthcare infrastructure across regions.

Regarding HIV/TB co-infection, 36.8% of the participants were co-infected, aligning with findings from a systematic review by Gelaw YA et al. (2019) in Sub-Saharan Africa<sup>16</sup>. This prevalence reflects the persistent burden of dual infection in high-risk populations. Similar studies have reported varying HIV/TB co-infection rates<sup>12-16</sup>, highlighting the strong link between the two diseases, particularly in regions with high HIV prevalence, where immune suppression accelerates TB progression. This underscores the need for integrated prevention, diagnosis, and treatment



strategies. However, studies by Masenga et al. (2017), Monde et al. (2021), and Chanda (2024) did not find a significant correlation between HIV and RR-TB<sup>3,4,15</sup>.

Our study observed the significant rise in RR-TB prevalence from 1.53% in 2020 to 4.6% in 2022 coincided with the COVID-19 pandemic, which disrupted TB services across Zambia. While overall TB prevalence remained relatively stable (11.41% in 2020 to 12.57% in 2022), the increasing trend in RR-TB cases highlights potential gaps in TB control efforts. Although the chi-square test for trend in RR-TB ( $\chi^2 = 4.82$ ,  $p = 0.09$ ) was not statistically significant likely due to the small sample size ( $n=20$ ), the observed pattern aligns with global reports of pandemic-related setbacks in TB management. This observed trend warrants further investigation, as it may reflect underlying challenges in TB control efforts, particularly in the context of the COVID-19 pandemic.

The rise in RR-TB cases aligns with global reports of disrupted TB services during the pandemic. Prior to COVID-19, TB control efforts had achieved significant progress, with steady declines in TB cases worldwide due to improved diagnostics, treatment strategies, and public health interventions<sup>17-20</sup>. However, the pandemic disrupted this trend, leading to a concerning resurgence in TB cases, including RR-TB<sup>19</sup>. In Zambia like many other resource limited settings, lockdowns and movement restrictions hindered access to healthcare facilities, leading to delays in TB diagnosis and interruptions in treatment. Many hospitals and clinics shifted resources toward COVID-19 response efforts, reducing TB testing capacity. Additionally, symptom overlap between TB and COVID-19 may have led to misdiagnosis or delayed case identification, allowing undiagnosed TB including RR-TB to spread within communities<sup>19,21</sup>. Studies have shown that incomplete or irregular TB treatment is a key driver of drug resistance, and the disruptions caused by the pandemic likely exacerbated this issue.

The rise in RR-TB is particularly alarming in Zambia, as it indicates the spread of drug-resistant strains of TB, which are more challenging and expensive to treat<sup>22</sup>. The COVID-19 disruptions likely contributed to the development and transmission of drug-resistant TB, as incomplete or interrupted treatment is a known driver of resistance<sup>19</sup>. This finding underscores the need for reinforced TB control strategies, including restoring diagnostic capacity, improving patient follow-up, and ensuring continuous access to TB medications. Strengthening surveillance systems to monitor emerging resistance patterns is also critical to mitigating the long-term impact of COVID-19 on TB management in Zambia.

The rising RR-TB prevalence despite stable TB rates underscores the need to update diagnostic guidelines. COVID-19-related delays, misdiagnosis due to overlapping symptoms, and resource constraints highlight gaps in current protocols. Policymakers should prioritize integrating rapid diagnostic tools like GeneXpert MTB/RIF into routine screening, enhance TB/HIV co-infection management, ensuring uninterrupted access to second-line treatments, and strengthen healthcare provider training. Improving sample referral systems and ensuring resource availability in high-burden areas will enhance early detection and treatment. Updating national TB control programs accordingly can help curb the spread of RR-TB and improve overall TB management.

Revisiting and updating diagnostic guidelines are essential to addressing the shortcomings exposed during the pandemic, improving early detection, and strengthening TB control efforts in Zambia. Enhancing these protocols will not only reduce the burden of drug-resistant TB but also ensure resilience against future health crises, mitigating the risk of RR-TB resurgence in the event of another pandemic like COVID-19.

## Limitations

The small RR-TB sample size (n=20) significantly constrained the depth of our analysis, particularly in subgroup comparisons, thereby limiting the statistical power to detect meaningful differences. To mitigate this, we combined age groups, but the findings should still be interpreted with caution. A larger sample would provide more robust conclusions and allow for more precise estimates of RR-TB prevalence across different demographics.

Additionally, the retrospective study design inherently limits the ability to establish causality. While we observed an increasing trend in RR-TB prevalence from 1.53% in 2020 to 4.6% in 2022, we cannot definitively attribute this rise to specific factors such as treatment failures, increased transmission of resistant strains, or improved diagnostic capacity. Retrospective data rely on existing records, which may lack crucial details on patient history, healthcare access, and environmental or behavioural risk factors. Prospective studies with comprehensive data collection on treatment adherence, prior TB exposure, and socioeconomic determinants would be necessary to establish causal relationships and better inform targeted TB control interventions.

## CONCLUSION

This study highlights the rising prevalence of rifampicin-resistant tuberculosis (RR-TB) at Ndola Teaching Hospital, Zambia, from 2020 to 2022, despite the stable overall TB rates. The increase in RR-TB, particularly among males and young adults, underscores the need for enhanced diagnostic capacity, targeted interventions for high-risk groups, and integrated HIV/TB care. The disruptions caused by COVID-19 further emphasize the importance of resilient health systems to withstand future crises. Strengthening surveillance, improving access to second-line treatments, and updating national TB control programs are essential to curb drug resistance and advance global TB elimination goals. Collaborative

efforts among researchers, healthcare providers, and policymakers are crucial to address these challenges effectively.

## What is already known on this topic:

Rifampicin-resistant tuberculosis is a major public health concern in Zambia, closely linked to multidrug-resistant TB (MDR-TB), which poses significant challenges for diagnosis and treatment. The persistence of rifampicin resistance threatens TB control efforts, making it a critical issue for healthcare systems. Studies have shown varying TB prevalence and resistance patterns across different regions, with HIV/TB co-infection remaining a significant burden. Additionally, the disruptions caused by the COVID-19 pandemic have further exacerbated TB management challenges, potentially contributing to the increasing prevalence of rifampicin-resistant TB.

## What this study adds:

This study provides valuable insights into the prevalence and trends of rifampicin-resistant tuberculosis at Ndola Teaching Hospital over a three-year period. It highlights a concerning increase in rifampicin resistance, despite a relatively stable overall TB prevalence. The study also underscores the high burden of TB among males and young adults and reveals a significant HIV/TB co-infection.

## Competing interests

The authors have no conflicts of interest to declare.

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Not applicable

## Authors' contributions

DC conceived the study. CM, NM and EC conducted data collection. DC and KMT conducted data analysis and writing of the manuscript. KMT and EC reviewed. All authors read and approved the final manuscript.

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