WOLKITE UNIVERSITY

COLLEGE OF MEDICINE AND HEALTH SCIENCES

DEPARTMENT OF MEDICAL LABORATORY SCIENCE

TRENDS OF MYCOBACTERIUM TUBERCULOSIS AND RIFAMPICIN RESISTANCE AT WOLKITE HEALTH CENTER, GURAGE ZONE, SOUTHERN ETHIOPIA

BY; ABATE TILAHUN

TRHAS HABTE

MARUFA MUSHKE

A RESEARCH PAPER SUBMITTED TO WOLKITE UNIVERSITY, COLLEGE OF MEDICINE AND HEALH SCIENCE, DEPARTMENT OF MEDICAL LABORATORY SCIENCE FOR A PARTIAL FULLFILMENT OF THE REQUIREMENTS FOR BACHELOR OF SCIENCE DEGREE IN MEDICAL LABORATORY SCIENCE

AUGUST, 2023 GC.

WOLKITE ETHIOPIA

Trends of Mycobacterium tuberculosis and rifampicin resistance at Wolkite health center, Gurage zone, Southern Ethiopia: Institutional Based Retrospective Study

BY; ABATE TILAHUN……0004/12

TRHAS HABTE……..…1458/12

MARUFA MUSHKE……..…0946/12

ADVISORS; Mr. REBIE KEDIR (BSC, MSC)

Mr. ASNAKE SEMENEH (BSC, MSC)

AUGUST, 2023 GC.

WOLKITE ETHIOPIA

ACKNOWLEDGMENT

We would like to acknowledge Wolkite University, College of medicine and health science, department of medical laboratory sciences for giving us this golden chance to develop this research paper.

We wish to express our sincere thanks, gratitude and appreciation to our advisors Mr. Rebie and Mr. Asnake for their enormous critical reading, valued advice, intellectual guidance and motivation for our research development. Finally, our thanks go to all neighboring friends for their constructive comments and suggestion during doing of this research.

Table of contents

Contents pages

List of tables

ABBREVIATIONS AND ACRONYM

AFB - Acid Fast Bacilli

AIDS - Acquired Immune Deficiency syndrome

DOTS - Direct Observation treatment short course

FMOH - Federal Ministry of Health

HIV - Human Immune Deficiency Virus

MDRTB - Multi-Drug Resistance Tuberculosis

MTB - Mycobacterium Tuberculosis

PTB - Pulmonary Tuberculosis

RIF Rifampicin

RR-MTB Rifampicin-resistant Mycobacterium tuberculosis

TB - Tuberculosis

WHO - World Health Organization

ABSTRACT

Background: Tuberculosis (TB) is one of the top ten causes of mortality and the first killer among infectious diseases worldwide. Drug resistance in TB remains a man-made phenomenon. It emerges as a result of spontaneous gene mutations in *M. tuberculosis* that render the bacteria resistant to the most commonly used anti-TB drugs, is a major global health problem. Rapid detection, earlier treatment initiation, continuous surveillance, and regular monitoring of drug-resistant TB are essential for disease management and control program.

Objective: To assess trends of Mycobacterium tuberculosis and rifampicin resistance in Wolkite health center from 2020 – 2023 G.C.

Methods: Retrospective study was conducted from June to August 15, 2023 GC. to determine trend analysis of prevalence of Mycobacterium tuberculosis and rifampicin resistance in the last 4 years at Wolkite health center in Gurage zone. Data collection was done from laboratory registration logbook by using standard checklist. The data was entered and analyzed by using SPSS statistical software package. Descriptive summary values such as frequency and percentage was presented using tables and graphs.

Result: From the total of 2411participants, the overall prevalence of MTB was 19.2% (462/2411). High prevalence was observed among male participants which, accountes about 294(63.6%). TB was reported in higher rate in the age groups 16-30 years, with a prevalence rate of 166(35.9%) followed by 31-45 years with a prevalence rate of 129(27.9%). From the total TB positive participants about, 47(10.17%) were Rifampin resistant. In each year from 2020 to 2023 EC, the prevalence of pulmonary TB was 16.78% (115/686), 17.85% (118/661), 20.65% (127/615) and 22.7% (102/449) respectively. And MDR-TB (Rifampicin Resistance) was 12.17% (14/115), 10.17% (12/118)14.5% (18/127) and 2.94% (3/102) respectively.

Conclusion: In this study, the overall prevalence of MTB was 19.2% (462/2411) with slightly increasing in the last year. From the total TB positive participants about, 47(10.17%) were Rifampin resistant. Therefore, maximizing early visit to health institutions and detection of Mycobacterium tuberculosis and enhancing active health education and intervention for tuberculosis are forwarded to reduce the threat of disease.

Keywords: MTB: Mycobacterium Tuberculosis; MDR-TB: Multidrug Resistance Tuberculosis; RIF: Rifampicin; TB: Tuberculosis; WHO: World Health Organization

CHABTER ONE

1. INTRODUCTION

1.1. BACK GROUND

Mycobacterium tuberculosis (MTB) is the causative agent of tuberculosis (TB), a chronic infectious disease that can be spread via the air by infected individuals (e.g. by coughing). It is latently present in approximately one-third of the world's population. In terms of a single infection, TB is one of the top 10 global causes of mortality. In 2019, about 10 million people were infected with MTB [1].

MTB resistance to anti-TB drugs can be either primary or acquired. Primary drug resistance of MTB is a resistance that is observed among patients without prior exposure to anti-TB drugs, while acquired drug resistance is a resistance observed among previously treated TB patients [2, 3]. Rifampicin is one of the most powerful anti-TB drugs used for the management of TB [4].

Drug resistance in TB remains a man-made phenomenon. It emerges as a result of spontaneous gene mutations in *M. tuberculosis* that render the bacteria resistant to the most commonly used anti-TB drugs. Among the reasons for this, the non-compliance with the treatment regimens is signaled as the first cause [5]. The majority of rifampicin-resistant MTB (RR-MTB) strains are mutated in the rpoB gene, which produces the RNA polymerase –subunit [6]. Most RR-MTB strains have mutations in 81 base pairs of the rpoB gene, which span codons 507 to 533 [7]

Rifampicin resistance has been utilized as a successful surrogate marker for multidrug-resistant MTB (MDR-MTB). Following the World Health Organization (WHO) guideline issued in 2017, all cases of RR-MTB, including those with MDR-TB, should be treated with second-line MDR-TB treatment regimens, about half million new cases of RR-MTB were reported among this, 78% were infected with MDR-MTB [8].

Emergence and spread of drug-resistant MTB are a challenge in the African region, especially in sub-Saharan Africa where TB control is difficult due to poor health infrastructure, limited resources, and lack of awareness [9]. Data on the burden of MDR-MTB from Africa are limited

due to lack of laboratory facilities, poor surveillance mechanisms, poor reporting procedures, and outdated database system [10]. About 60,000 MDR-MTB cases occur annually in the sub-Saharan region, which represents about 14% of the world's MDR-MTB burden [11]. Ethiopia stands 15th out of the 27 high-priority countries in the world and 3rd in Africa next to South Africa and Nigeria [12]. According to the second survey conducted in Ethiopia, the prevalence of RR-MTB in Ethiopia was lower than 2% [13]. In 2016, in Ethiopia, there were 219,186 new and 156,602 prevalent TB cases and 48910 TB-related deaths [14].

Drug-resistant MTB can be controlled by identifying and effectively treating sputum smear-positive cases. Furthermore, prophylaxis and health education have an essential role in controlling the spread of MDR-TB [15]. The WHO recommends DOTS (direct observation therapy short-course) and the Stop TB strategies to address the needs of the poor and vulnerable population [16].Understanding the drug susceptibility patterns of *M. tuberculosis* (MTB) is very crucial to treat patients, to decide health priorities, to allocate resources, to monitor the emergency of resistance for planning effective use of anti-TB drugs [17].

Unless individuals infected with drug-resistant MTB are treated appropriately, they will continue disseminating MTB in the community and accelerate the epidemics. The impact of MDR-MTB is serious especially in low-income countries like Ethiopia, where health resources, finances, and the number of skilled personnel are limited [18]. Although more advanced methods for diagnosing TB, like fluorescence microscopy and Gene X-pert (Cepheid), are being used in Ethiopia, acid fast Bacilli (AFB) is still the primary method in the majority of the nation.

Southern Nations, Nationalities, and People's Region (SNNPR) has a population of more than 18 million, and more than 90% of the population lives in rural communities. Whereas diagnosis and treatment of TB are carried out in hospitals and health centers, directly observed therapy (DOT) is decentralized to health centers in SNNPR. Furthermore, at the community level, DOT is provided at the health posts. A study by Datiko et al reported 960 presumptive TB cases from a total of 38,304 studied subjects in three zones of SNNPR (Hadiya, Gurage, and Sidama). The point prevalence (smear-positive) of TB cases from Hadiya, Gurage, and Sidama zones (now Sidama region) was 148, 139, and 80 per 100,000 populations respectively [19].

If MTB from TB suspected patients becomes resistant to rifampicin by Gene X-pert, they will be considered as drug-resistant (DR-TB) and referred to the MDR-TB treatment initiative center

(TIC). Then, the patient will be admitted to the TIC and the sputum specimens will be collected and referred to the regional laboratory for confirmation and monitoring using culture method, line probe assay (LPS), microscopy, and drug sensitivity tests. A standard second-line treatment will be given to all MDR/RR-confirmed cases daily under direct observation by a health care worker at a health care center and family DOT supporter(s) at home. The regimens include at least four second-line anti-TB drugs that are certain or expected to be effective, and the duration is a minimum of 18 months after culture conversion [20].

1.2. STATEMENT OF THE PROBLEM

Tuberculosis (TB) is a major cause of ill health, one of the top 10 causes of death worldwide and the leading cause of death from a single infectious agent (ranking above HIV/AIDS since 2007). Globally in 2019, an estimated 10.0 million (range, 8.9–11.0 million) people fell ill with TB. There were 1.2 million (range, 1.1–1.3 million) TB deaths among HIV-negative people and an additional 208 000 deaths (range, 177 000–242 000) among HIV-positive people [4].

TB affects people of both sexes and all age groups, but the highest burden is in adult men, who accounted for 56% of all TB cases in 2019; by comparison, adult women accounted for 32% and children for 12%. Among all TB cases, 8.2% were among people living with HIV. Geographically, in 2019, most TB cases were in the World Health Organization (WHO) regions of South-East Asia (44%), Africa (25%) and the Western Pacific (18%), with smaller shares in the Eastern Mediterranean (8.2%), the Americas (2.9%) and Europe (2.5%). Eight countries accounted for two thirds of the global total: India (26%), Indonesia (8.5%), China (8.4%), the Philippines (6.0%), Pakistan (5.7%), Nigeria (4.4%), Bangladesh (3.6%) and South Africa (3.6%) [4].

Out of the total TB incident cases reported in 2018, 484,000 were resistant to rifampicin (RR-TB), and of these, 78% (3.4% new cases and 18% previously treated cases) had multidrug-resistant TB (MDR-TB). A total of 214,000 patients died due to MDR/RR-TB in 2018 [22].

In 2018, an estimated 3.4% of the global TB cases were new drug resistant (MDR/RR) and 18% were among previously treated cases. In Ethiopia, the estimated incidence of MDR/RR is 0.71 and 16% among new and previously treated cases, respectively [22]. In addition, the estimated cases of MDR-RR had a magnitude of 484,000, which was about 10% downward from the best estimate published by WHO in its 2018 global TB report. Of these estimated cases, about 44.2% (214,000) deaths were due to MDR/RR-TB, which was also a downward revision to the best estimates. In fact, the global notified cases rather than estimates of MDR/RR-TB were 186, 772 up from 160,772 cases in 2017, and 156,071 cases were enrolled in treatment which was also up from 139, 114 in 2017. The number of people enrolled in treatment in the year was equivalent to only 32% of the estimated incidence of the 484,000 cases [22, 23].

Several studies in Ethiopia assessed the magnitude of drug-resistant TB. A review done by Biadglegne et al. reported that the occurrence of MDR-TB among TB patients in Ethiopia ranged

from 3.3% to 46.3%. Moreover, based on a recent meta-analysis report, the pooled estimate of MDR-TB among new and previously treated cases was 2% (1 to 2%) and 15% (12 to 17%), respectively. Another study reported a MDR-TB prevalence ranging from 0 to 46.3% [23].

Ethiopia is among the 30 high TB, TB/HIV, and MDR-TB burdened countries with a rank of 15th among high MDR-TB countries with more than 5800 estimated MDR-TB cases each year [27]. A systematic review conducted in Ethiopia reported 2.18% and 21.07% prevalence of MRDTB among new and pre-treatment cases, respectively [25].

The early diagnosis and treatment of TB patients is mandatory to reduce transmission of the disease. Millions of people are diagnosed and successfully treated for TB each year, averting millions of deaths, but there are still large gaps in detection and treatment. For the application of control policy, efforts for the identification of TB cases and treatment are mandatory which is not currently sufficient. Because of this, updated knowledge of the prevalence of M. tuberculosis and drug resistance is crucial. Hence, the present study will be intended to provide updated information on the prevalence of mycobacterium tuberculosis and rifampicin resistance at Wolkite health center, Southern Ethiopia.

1.3. SIGNIFICANCE OF THE STUDY

The result of this study was supposed to provide information on the trend analysis of prevalence of mycobacterium tuberculosis and rifampicin resistance to Wolkite health center laboratory. It will also give information to Gurage zone health bureau and other stakeholder in order to develop workable intervention for controlling the transmission pulmonary tuberculosis. In addition, it will provide base line information to those who are interested for further study on prevalence of mycobacterium tuberculosis and rifampicin resistance.

CHAPTER TWO

2. LITERATURE REVIEW

2.1. Prevalence of Tuberculosis and Rifampicin resistance

Tuberculosis (TB) continues to be a major public health problem in the world. In 2021, eight countries accounted for more than two thirds of global TB cases: India (28%), Indonesia (9.2%), China (7.4%), the Philippines (7.0%), Pakistan (5.8%), Nigeria (4.4%), Bangladesh (3.6%) and Democratic Republic of the Congo (2.9%) [26]

Globally, the burden of multidrug- or rifampicin-resistant TB (MDR/RR-TB) as a share of the number of TB cases remains stable. In 2019, an estimated 3.3% of new TB cases and 18% of previously treated cases had MDR/RRTB, in absolute numbers, there were an estimated 465 000 (range, 400 000– 535 000) incident cases of rifampicin resistant TB; India (27%), China (14%) and the Russian Federation (8%) had the largest share of the global burden [4].

Tuberculosis (TB) continues to be a major public health problem in Ethiopia. In 2020, of the 157,000 estimated cases of TB, 108 of 193 (71 percent) were notified, among which 70 percent were pulmonary TB cases and only 62 percent of these pulmonary TB cases were bacteriologically confirmed [21].

TB is the most common cause of morbidity and mortality in Ethiopia. The country is among the three highest TB, TB/HIV and MDR-TB burden countries with estimates of 165,000 new TB cases and a rate of 151/100,000 population reported in 2018. In the same year, the number of cases of MDR/RR-TB was 1,600; the number of fatalities from TB was 24,000 for HIV-negative people and an additional 2,200 when including people living with HIV/ AIDS [22].

A study conducted in northern Ethiopia indicates of 124 smear-positive pulmonary TB patients, 117 (94.4 %) were susceptible to Rifampicin, while 7 (5.7 %) were confirmed to be resistant to Rifampicin and Isoniazid. The overall prevalence of MDR-TB was 5.7 % (2.3 % among new cases and 13.9 % among previously treated cases) [17].

A retrospective study conducted at Tigray in 2019 shows that a total of 7793 tuberculosis presumptive clients were requested for laboratory diagnosis of which about 7639 results had a

valid result for X-pert MTB/Rif assay. The overall detection rate of tuberculosis was found to be 9.9% (756/7639). Of the total tuberculosis cases, 8.7 % (66/756) were rifampicin-resistant [27].

A cross-sectional study conducted at Debre Markos Referral Hospital from September 2014 to March 2015 that Detection of M. tuberculosis and resistance to rifampicin was performed using Gene X-pert MTB/RIF assay show that, from a total of 505 presumptive TB patients, the prevalence of M. tuberculosis confirmed cases was 117 (23.2%) (95 % CI 19. 7 – 27 %), of the 117 M. tuberculosis confirmed cases, 12 (10.3%) (95% CI 6.0–17.1%) were resistant to rifampicin. Rifampicin-resistant M. tuberculosis was noticed in 7 previously treated TB patients (17.1%) and 5 treatment naive patients. [39].

A retrospective cross sectional study conducted in three referral hospitals and regional laboratory in Addis Ababa city from March 2015 to October 2017 show that, from the total of 12,414 (11,672 adults and 742 pediatrics) TB presumptive patients, the overall prevalence of TB was 15.11% (1876/12414) in all age groups and 13.6% (101/742) among pediatric population. Rifampicin resistant TB was 9.9% (186/1876) in all TB confirmed cases and 7.9% (8/101) in pediatric TB patients. The prevalence of rifampicin resistant TB among new and previously treated was 7.6 and 27.4%, respectively [28].

A hospital-based cross-sectional study conducted at Adare General Hospital located in Hawassa city from April to July 2018 show that, from the total 321 tuberculosis suspected patients, the prevalence of Mycobacterium tuberculosis was 98 (30.5%), From total of confirmed TB cases 4 (4.1%) patients developed rifampicin resistance [20].

A health institution-based retrospective cross-sectional study conducted at Wolkite town Health center from January 2016 to 2020 show that from the total of 3286 participants of complete Gene X-pert MTB/Rif assay, The overall detection rate of TB was 27.3 % (899/3286) and the prevalence of TB was higher in males 500 (55.6%) than females 399 (43.4%). From total of confirmed TB cases, 137 (15.2%) participants developed rifampicin resistance [29].

CHAPTER-THREE

3. OBJECTIVES

3.1. General Objective

- To assess trends of *Mycobacterium tuberculosis* and its rifampicin resistance at Wolkite health center in Gurage one from 2020 – 2023 G.C.

3.2 Specific objective

- To assess the four-year trend of *Mycobacterium tuberculosis* at Wolkite health center
- To assess the trend of rifampicin resistant tuberculosis at Wolkite health center.

CHAPTER-FOUR

4. METHODS AND MATERIALS

4.1. Study area

The study was conducted in Wolkite town health center, located in Gurage Zone. Wolkite town is located 158 km southwest of Addis Ababa along the Jimma Road in the Southwest Region of Ethiopia. Wolkite town, the capital city of Gurage Zone, has a latitude and longitude of 8°17'N37°47'E, an average annual temperature of 18.6 °C and an average rainfall of 1244 mm. The town has an elevation between 1910 and 1935 meters above sea level. Based on the 2007 Census conducted by the Central Statistical Agency of Ethiopia, Wolkite town has a total population of 28,856 of whom 15,068 were males and 13,788 females. Wolkite health center offers diagnosis and treatment for patients that reside in Wolkite town and nearby neighbor woredas like Kebena, Abeshge and others.

4.2. Study Design and study period

A health institution-based retrospective study was conducted from June to August 15, 2023 G.C. to assess the trend analysis on prevalence of mycobacterium Tuberculosis and rifampicin resistance.

4.3. Study Population

Those presumptive tuberculosis patients who visited Wolkite health center TB laboratory and recorded on Gene X-pert TB registration logbook from January 2020 – June 2023 G.C.

4.4. Sample size

A total of 2411 presumptive TB clients were examined for X-pert MTB/Rif assay from different areas in Wolkite health center.

4.5. Variables

- Tuberculosis
- Age
- Address
- Reason for Dx
- Sex
- Year of diagnosis Seasons

4.6. Eligibility criteria

4.6.1. Inclusion criteria

Data from 2411 MTB suspected patients with complete results in the last four years were included.

4.6.2. Exclusion Criteria

321 Patients with incomplete information on Gene X-pert TB registration logbook excluded from the analysis

4.7. Operational Definitions

New case (N): a patient who never had treatment for TB or has been on previous anti TB treatment for less than four weeks.

Relapse(R): are previously treated for TB, were declared cured or treatments completed at the end of their most recent treatment episode and are now diagnosed with a recurrent episode of TB.

Treatment Failure (TF): are previously treated for TB and whose treatment failed at the end of their most recent treatment episode.

Return after default (D): patient whose treatment was interrupted for two or more consecutive months for any non-medically approved reason and returns to health facility with smear positive sputum.

MDR TB: a form of TB caused by bacteria that do not respond to, at least, isoniazid and rifampicin, the two most powerful, first-line (or standard) anti-TB drugs.

RTB: recurrent tuberculosis pulmonary tuberculosis patients who had previously been treated and declared as cured prior to becoming once again bacteriologically positive.

PTB: pulmonary TB type of TB caused by mycobacterium tuberculosis which affect lung mainly.

4.8. Data collection and Analysis

Data was collected with predesigned checklist from the registration logbook. The collected data include age, sex, and date of diagnosis and the result of Gene X-pert test. The data was entered and analyzed by using SPSS statistical software package.

4.9. Data Quality Control

Data was checked for consistency and completeness, and incomplete data was excluded from the analysis. Secondary data was obtained appropriately from registration logbook of Gene X-pert result.

4.10. Ethical consideration

The study was approved by college of medicine and health sciences, institutional research ethical committee, Wolkite University. Then formal letter of cooperation was written to Wolkite health center administration. The objective of the study was explained to the health center. The letter of agreement was attached to ensure confidentiality of data. The information taken from patients' recorded data was kept confidential. Only codes were used to identify the study groups.

4.11. Plans for dissemination of finding

The result of the study was appropriately communicated to the health center and representative of the department. Our result was disseminated to Wolkite University College of medicine and health science, department of medical laboratory sciences for further investigation by senior researchers. In addition to this, our result will disseminate into Gurage health bureau for further research and to take measure by creating awareness in the prevalence of disease.

CHAPTER FIVE

5. Result

5.1 Socio-demographic and clinical data of Study Subject

From the total of 2732 TB clinically suspected patients that have submitted their sputum samples for TB diagnosis in four year, 2411 have complete data in the registration logbooks. From the total study participants, 1397(57.9%) were males and 696(28.9%) study participants were found in age group of 31-45 followed by age group 46-60 and 16-30 which accounted 686 (28.5%) and 625 (25.9%) respectively.

Majority of study participants were presumptive TB case 2380 (98.7%). Among total study participants 2249 (93.3%), were N (New case) and 146 (6.1%), were R (Relapse) and the remaining 9(0.4%). and 7(0.3%) were AF (After failure of first treatment) and TA (Treatment after lost to follow up) respectively in patient registration group. The number TB suspected patients diagnosis for Mycobacterium tuberculosis using the Gene X-pert™ have decreased gradually from 2020 to 2023. From the total of study participants only 176 (7.3%) know their HIV status, from which 78 (44.3%) were living with HIV. (Table 1)

Table 1: Socio-demographic characteristics of study participants

Variables		Frequency	Percent (%)
Sex	M (Male)	1397	57.9
	F (Female)	1014	42.1
Age	<5	22	0.9
	5-15	113	4.7
	16-30	625	25.9
	31-45	696	28.9

	46-60	686	28.5
	>60	269	11.2
HIV Status	P (Positive)	78	3.2
	N (Negative)	98	4.1
	Un (Unknown)	2235	92.7
Patient Residence	Wolkite	742	30.8
	Cheha	274	11.4
	Abeshge	283	11.7
	Kebena	122	5.1
	Wolen	120	5
	Gumer	125	5.2
	Atat	147	6.1
	Other	598	24.8
Patient Registration Group	N (New case)	2249	93.3
	R (Relapse)	146	6.1
	TA (Treatment after lost to follow up)	7	0.3
	AF (After failure of first treatment)	9	0.4

5.2. Prevalence of tuberculosis and rifampicin resistance

From the total of 2411 study participants diagnosed within the last 4-years, 462(19.2%) was contracted by Mycobacterium tuberculosis infection. Higher prevalence of tuberculosis was observed among male accounted 294(63.6%) than female 168(36.4%). TB was reported in all age groups but the infection rate was higher in the age groups 16-30 years, with a prevalence rate of 166(35.9%) followed by 31-45 and 46-60 age groups with a prevalence rate of 129(27.9%) and 119(25.8%) respectively. Among the MTB positives 11(2.4%) were HIV positive, 16(3.5%) were negative and the rest 435(94.2%) were unknown HIV status. From the total confirmed all forms of Mycobacterium tuberculosis cases, 93.3% (431/462) were identified from presumptive TB and 6.7% (31/462) from presumptive drug resistance-TB.(Table 2)

Table 2: prevalence of TB with in Socio-demographic and Clinical Characteristics

Variables		TB Status	
		P (%)	N (%)
Sex	M	294(63.6)	1103 (56.6)
	F	168(36.4)	846(43.4)
Age(years)	<5	2(0.4)	20 (1.0)
	5 – 15	6 (1.3)	107 (5.5)
	16 – 30	166 (35.9)	459 (23.5)
	31 – 45	129 (27.9)	567 (29.1)
	46-60	119 (25.8)	567 (29.1)
	>60	40 (8.7)	229 (11.7)
Patient Residence	Wolkite	131 (28.4)	611 (31.3)
	Cheha	63 (13.6)	211 (10.8)

	Abeshge	58 (12.6)	225 (11.5)
	Kebena	25 (5.4)	97 (5)
	Wolen	23 (5)	97 (5)
	Gumer	27 (5.8)	98 (5)
	Atat	29 (6.3)	118 (6.1)
HIV Status	P	462 (100)	1949 (100)
	N	16(3.5)	82(4.2)
	Un	435(94.2)	1800(92.4)
Patient Registration Group	NC	336 (72.7)	1913 (98.2)
	R	114 (24.7)	32 (1.6)
	TA	5 (1.1)	2 (0.1)
	AF	7 (1.5)	2 (0.1)

Key words: M – Male, F – Female, Un (Unknown), N – Negative, P – Positive, NC – New Case, R – Relapse, TA – Treatment after lost to follow up, AF – After failure of first treatment

5.3. Prevalence of Rifampin Resistance Tuberculosis

From the total TB positive cases, about 47(10.17%) were Rifampin resistant tuberculosis. MDR-TB was reported in age groups 31-45, 46-60, 16-30 and >60 with a prevalence rate of 15(31.9%), 15(31.9%), 11(23.4%) and 6(12.8%) respectively. Among the positive 4(8.5%) was HIV positive, 1(2.1%) was HIV negative and 42(89.4%) was unknown HIV status. Among 47 patients who are positively examined in the past four years the patient registration group was new cases 16(34%), relapse 29(61.7%) and after failure of first treatment2(4.3%). (Table 3)

Table 3: prevalence of Multi-Drug Resistance TB with Socio-Demographic and Clinical Characteristics Patient

Variables		RR Status	
		Sen (%)	Res (%)
Sex	M	260 (62.65)	34 (72.3)
	F	155 (37.35)	13 (27.7)
Age	<5	2 (0.4)	0 (0)
	5-15	6 (1.4)	0 (0)
	16-30	155 (37.3)	11 (23.4)
	31 – 45	114 (27.4)	15 (31.9)
	45 – 60	104 (25.06)	15 (31.9)
	>60	34 (8.1)	6 (12.8)
HIV Status	P	7 (1.6)	4 (8.5)
	N	15 (3.6)	1 (2.1)
	Un	393 (94.6)	42 (89.4)
Patient Registration Group	NC	320 (77.1)	16 (34)
	R	85 (20.4)	29 (61.7)
	AF	5 (1.2)	2 (4.3)
	TA	5 (1.2)	0(0)

Key words: M – Male, F – Female, Un (Unknown), Sen – Sensitive, N – Negative, P – Positive, NC – New Case, R – Relapse, AF – After failure of first treatment, TA – Treatment after lost to follow up

5.4. Four-year Trend of Tuberculosis and Rifampicin resistant tuberculosis

In each year from 2020 to 2023, the prevalence of pulmonary TB was 16.78% (115/686), 17.85% (118/661), 20.65% (127/615) and 22.7% (102/449) respectively. And MDR-TB (Rifampicin Resistance) was 12.17% (14/115), 10.17% (12/118)14.5% (18/127) and 2.94% (3/102) respectively. The overall prevalence of TB in the Four years had an increasing trend. (Table 4)

Table 4: Trend of Tuberculosis and Multi-Drug Resistance Tuberculosis

Variables		TB Status		Total	Rif. Resistance	
		N (%)	P (%)		Sen (%)	Res (%)
Years	2020	571(83.23)	115(16.78)	686(28.5%)	101(87.8)	14(12.17)
	2021	543(82.14)	118(17.85)	661(27.4%)	106(89.8)	12(10.17)
	2022	488(79.34)	127(20.65)	615(25.5%)	107(84.25)	18(14.5)
	2023	347(77.28)	102(22.7)	449(18.6%)	89(87.25)	3(2.94)
	Total	1949(80.8)	462(19.2)	2411(100%)	403(87.2)	47(10.17)

Keywords: N- Negative, P- Positive, Sen- Sensitive and res-resistance

CHAPTER SIX

6. Discussion

In this study, four-year retrospective data collected to determine the trend prevalence of mycobacterium tuberculosis among clients visited to TB clinic of Wolkite town heath center. The overall detection rate of TB in the study was 19.2% and MDR-TB was 10.17%. The prevalence of TB was higher among people of productive age groups 16–30 and 31-45 years. In the years from 2020 to 2023, the prevalence of TB were 115 (24.9%), 118 (25.5%), 127 (27.5%) and 102 (22.1%) respectively. Our finding was comparable with study report by WHO in Africa 25% [4], Southern Sierra Leone 21.3% [30], Nigeria 22.9% [31], Eastern Ethiopia 15.7% [32], Afar (24.5%) [33], Eastern zone of Tigray 24.3% [34], Debre Markos Hospital 23.2% [39] and Gambella 20.0% [35].

However, our study showed higher overall Mycobacterium tuberculosis prevalence than other studies conducted Northwestern Tigrai, Ethiopia 9.9% [27], Debre Berhan, Ethiopia 13% [36], Felege Hiwot Referral Hospital and Debre Tabor Hospitals 14.6% [37], Addis Ababa 15.11% [28], Indonesia 9.2% [26]. In contrary, our study revealed lower prevalence of mycobacterium tuberculosis compared to other studies conducted in wolkite health center 27.3% [29], Adare general hospital in Hawassa 30.5% [20] Jigjiga, Ethiopia 65.5% [38]and India 28% [26]. This difference might be due difference in the method of diagnosis, duration of study period, community characteristics and geographical location, skill of the laboratory personnel to detect TB, study design and other factors that may affect case occurrences in different study areas.

The rate of rifampicin resistance was 10.17% in our study among confirmed TB cases. This result was agreed with studies done in Debre Markos Referral Hospital 10.3% [39], FelegeHiwot Referral Hospital and Debre Tabor Hospital 9.3% [37], Addis Ababa 9.9% [28], Eastern zone of Tigrai, Ethiopia 9.1% [34] and India 14% [4]. However, our finding was higher than the study done in Tigray 8.7% [27], northern Ethiopia 5.7% [17], Adare General Hospital Hawassa 4.1% [20] and Russia 8% [4]. But lower rifampicin resistance rate was reported in our study than studies done in wolkite health center 15.2% [29]. The possible reason for variations might be related to differences in study designs and drug utilization manner of patients. In our study Regarding the age group, 16-30 years were more affected by MTB 166 (35.9%) followed by 31-

45 years 129(27.9%). This was consistent with previous reports in different part of Ethiopia cited elsewhere above. This might be due to more exposure to the outer environment, high workload and wide range of mobility of young people to acquire the TB bacilli. In the current study, the Mycobacterium tuberculosis detection rate was higher in presumptive TB patients compared to presumptive drug resistance TB patients.

CHAPTER SEVEN

7. CONCLUSION AND RECOMMENDATION

7.1. Conclusion

The overall prevalence of MTB in wolkite health center was 19.2% (462/2411) with slightly increasing in the past four years (2020 - 2023). High prevalence was observed among male accounted 294(63.6%). TB was reported in higher rate in the age groups 16-30 years, with a prevalence rate of 166(35.9%) followed by 31-45 years with a prevalence rate of 129(27.9%). From the total study participants, 47(10.17%) were positively examined for Rifampin resistant tuberculosis from those 34(72.3%) was male and the rest 13(27.7%) was female. MDR-TB was reported in age groups 31-45, 46-60and 16-30 with a prevalence rate of 15(31.9%), 15(31.9%) and 11(23.4%) respectively.

7.2. Recommendation

In this study, the overall trends of Mycobacterium tuberculosis and rifampicin resistance were found to be significant and with variation each year. Rifampicin resistance is more common in presumptive drug resistance tuberculosis individuals. Therefore, maximizing early detection of drug-resistant and strengthening tuberculosis infection control activities are recommended to reduce the burden of this contagious and potentially deadly disease.

To tackle the effect of TB disease the respective health authorities, Wolkite health center, Gurage health bureau zone and governmental bodies should;

- Give health education about the character of the disease, mode of transmission and method of prevention in a continuous manner.
- Create awareness about the effect of associated risk factor such as occupational status, alcohol drinking
- Initiate further studies to identify clearly the risk factors for PTB and burden of the disease as well.

7.3. Limitation of the study

As retrospective data was collected from the Gene X-pert™ TB registration book, missing and incompleteness of data was faced.

6. REFERENCES

1. *Global tuberculosis report*. Geneva: World Health Organization, 2020,
2. Sharma SK, Mohan A. Multidrug-resistant tuberculosis. *Indian J Med Res* 2004; 120: 354–376.
3. Jain A, Dixit P. Multidrug resistant to extensively drug resistant tuberculosis: what is next? *J Biosci* 2008; 33(4): 605–616.
4. *Global tuberculosis report*. Geneva: World Health Organization, 2019,
5. Juan Carlos and Anandi Martin, Drug Resistance Mechanisms in Mycobacterium tuberculosis (2014)
6. Portelli S, Phelan JE, Ascher DB, et al. Understanding molecular consequences of putative drug resistant mutations in *Mycobacterium tuberculosis*. *Sci Rep* 2018; 8: 15356.
7. Hameed HMA, Islam MM, Chhotaray C, et al. Molecular targets related drug resistance mechanisms in MDR-, XDR-, and TDR-*Mycobacterium tuberculosis* strains. *Front Cell Infect Microbiol* 2018; 8: 114.
8. Ramaswamy S, Musser JM. Molecular genetic basis of antimicrobial agent resistance in Mycobacterium tuberculosis: 1998 update. *Tuber Lung Dis* 1998; 79(1): 3–29.
9. Lukoye D, Ssengooba W, Musisi K, et al. Variation and risk factors of drug resistant tuberculosis in sub-Saharan Africa: a systematic review and meta-analysis. *BMC Public Health* 2015; 15(1): 291.
10. Musa BM, Adamu AL, Galadanci NA, et al. Trends in prevalence of multi drug resistant tuberculosis in sub-Saharan Africa: a systematic review and metaanalysis. *PLoS ONE* 2015; 12(9): e0185105.
11. Wells CD, Cegielski JP, Nelson LJ, et al. HIV infection and multidrug—resistant tuberculosis—the perfect storm. *J Infect Dis* 2007; 196(Suppl. 1): S86–S107.
12. World Health Organization (WHO). *Global tuberculosis report*. Geneva: WHO Press, 2012,
13. World Health Organization (WHO). *Global tuberculosis report*. Geneva: World Health Organization, 2014,
14. Deribew A, Deribe K, Dejene T, et al. Tuberculosis burden in Ethiopia from 1990 to 2016: evidence from the Global Burden of Diseases 2016 study. *Ethiop J Health Sci* 2018; 28(5): 519–528.

15. Velayati AA, Farnia P, Mozafari M, et al. High prevalence of rifampin-mono resistant tuberculosis: a retrospective analysis among Iranian pulmonary tuberculosis patients. *Am J Trop Med Hyg* 2014; 90(1): 99–105.
16. Falzon D, Schünemann HJ, Harausz E, et al. World Health Organization treatment guidelines for drug-resistant tuberculosis, 2016 update. *EurRespir J* 2017; 49(3): 1602308.
17. Mekonnen F, Tessema B, Moges F, et al. Multidrug resistant tuberculosis: prevalence and risk factors in districts of Metema and West Armachiho, Northwest Ethiopia. *BMC Infect Dis* 2015; 15(1): 461.
18. Eshetie S, Gizachew M, Dagnew M, et al. Multidrug resistant tuberculosis in Ethiopian settings and its association with previous history of anti-tuberculosis treatment: a systematic review and meta-analysis. *BMC Infect Dis* 2017; 17(1): 219.
19. Datiko DG, Guracha EA, Michael E, et al. Sub-national prevalence survey of tuberculosis in rural communities of Ethiopia. *BMC Public Health* 2019; 19: 295.
20. DeginehBelachew, Tariku, GetamesayMulat and Musa Mohammed; (2018) Prevalence of Mycobacterium tuberculosis infection and rifampicin resistance among presumptive tuberculosis cases visiting tuberculosis clinic of Adare General Hospital, Southern Ethiopia
21. Ethiopia Tuberculosis Roadmap Overview, Fiscal Year 2022
22. World Health Organization. Global Tuberculosis report 2018 [Internet], Geneva; 2019,
23. Welekidan LN, Skjerve E, Dejene TA, Gebremichael MW, Brynildsrud O, Agdestein A, et al. (2020) Characteristics of pulmonary multi drug resistant tuberculosis patients in Tigray Region, Ethiopia: A cross-sectional study.
24. World Health Organization. Global tuberculosis report 2017. Geneva, Switzerland
25. Girum T, Muktar E, Lentiro K, Wondiye H, Shewangizaw M. Epidemiology of multidrug-resistant tuberculosis (MDR-TB) in Ethiopia: a systematic review and meta-analysis of the prevalence, determinants and treatment outcome.
26. WHO Global Tuberculosis Report 2022, December 12,2022DOI:
27. HadushNegash, HaftomLegese, GebreAdhanom, FitsumMardu, KebedeTesfay, SenaitGebreslasieGebremeskel and BrhaneBerhe; Six Years Trend Analysis of Tuberculosis in Northwestern Tigrai, Ethiopia; 2019: A Retrospective Study

28. BalewArega, FiqrteMenbere&YitagesuGetachew; (2019) Prevalence of rifampicin resistant *Mycobacterium tuberculosis* among presumptive tuberculosis patients in selected governmental hospitals in Addis Ababa, Ethiopia Arega et al. BMC Infectious Diseases (2019) 19:307
29. Admasu Haile, Kassahun Haile and SofiyaShemsu;(2021) Trend and Associated Factors of Mycobacterium Tuberculosis and Rifampicin Resistance in South-west Ethiopia: Institutional Based Retrospective Study
30. Babawo L, Sellu E, George A, Kaikai D (2020) A Five-Year Incidence Trend Analysis of Tuberculosis (TB) and HIV/AIDs Co-Infection at Bo Government Hospital, Southern Sierra Leone. J Public Health Dis Prev 3: 2
31. Ikuabe PO, Ebuenyi ID (2018) Prevalence of rifampicin resistance by automated Genexpert rifampicin assay in patients with pulmonary tuberculosis in Yenagoa, Nigeria. Pan Afr Med J 29.
32. Bodena D, Ataro Z, Tesfa T (2019) Trend analysis and seasonality of tuberculosis among patients at the HiwotFana Specialized University Hospital, Eastern Ethiopia: a Retrospective Study. Risk ManagHealthc Policy 12: 297
33. Gebrehiwet GB, Kahsay AG, Welekidan LN, Hagos AK, Abay GK, et al. (2019) Rifampicin resistant tuberculosis in presumptive pulmonary tuberculosis cases in Dubti Hospital, Afar, Ethiopia. J Infect DevCtries 13: 21-27
34. Abay GK, Abraha BH (2020) Trends of Mycobacterium tuberculosis and rifampicin resistance in Adigrat General Hospital, Eastern zone of Tigrai, North Ethiopia. Trop Dis Travel Med Vaccines 6: 1-9.
35. Ejeta E, Beyene G, Bonsa Z, Abebe G (2018) Xpert MTB/RIF assay for the diagnosis of Mycobacterium tuberculosis and Rifampicin resistance in high human immunodeficiency virus setting in gambella regional state, southwest ethiopia. J ClinTuberc Other Mycobact Dis 12: 14-20.
36. Asfaw T, Terefe A, Nigus M (2018) Patterns and Trends of RifampicinResistance Mycobacterium tuberculosis and Associated Factors among Presumptive Tuberculosis Patients at DebreBerhan Referral Hospital. J Bioprocess Biotech 8: 2.
37. Derbie A, Worku S, Mekonnen D, Mezgebu Y, Teshager A, et al. (2016) Xpert MTB/RIF assay for the diagnosis of Mycobacterium tuberculosis and its Rifampicin resistance at

FelegeHiwot and Debre Tabor Hospitals, Northwest Ethiopia: A preliminary implementation research. Ethiop J Health Dev 30: 60-66.

38. Worku M, Agonafir M, Yassin MA, Yassin MA, Datiko DG, et al. (2019) Use of Xpert MTB/RIF for the Identification of TB and Drug Resistance Among Smear-Negative and Re-Treatment Cases in Rural Areas of Ethiopia. Open Microbiol J 13
39. Mulu W, Abera B, Yimer M, et al. Rifampicin-resistance pattern of Mycobacterium tuberculosis and associated factors among presumptive tuberculosis patients referred to DebreMarkos Referral Hospital, Ethiopia: a cross-sectional study. *BMC Res Notes* 2017; 10(1): 8.

ANNEX- ONE

WOLKITE UNIVERSITY

COLLEGE OF MEDICINE AND HEALTH SCIENCE

DEPARTMENT OF MEDICAL LABORATORY SCIENCES

Checklist for data collection for the prevalence of mycobacterium tuberculosis and rifampicin resistance will be conducted among patients visited Wolkite health center in the past four years.

Code town number

1. Id N^{o}_____

2. Time of examined:

Year _____ Month: _____ Day_____

3. Sex:

☐ Male ☐ Female

4. Age: _____

5. Case

☐ New Case ☐ Relapse Case

6. Gene x-pert Results

☐ T ☐ RR ☐TI

☐N ☐NR ☐I ☐E

7. Type of registration group

i. New
ii. Relapse
iii. Treatment after lost to follow up
iv. after failure of first treatment

8. Defaulted HIV status

a. Positive b. Negative c. Unknown

9. Reason for examination 1.Diagnosis 2, presumptive T

ANNEX-TWO

2. Gene X-pert

What is usually referred to as the Gene x-pert test, has now been developed into a series of molecular tests. Depending on the cartridge/assay used, the Gene x-pert can test for a number of infectious diseases including TB & COVID. The Gene x-pert diagnoses TB by detecting the presence of TB bacteria, the test is a molecular TB test which detects the DNA in TB bacteria. It uses a sputum sample and can give a result in less than 2 hours. The Gene x-pert can also detect the genetic mutations associated with resistance to the drug Rifampicin.

The Gene x-pert has been developed by the Foundation for Innovative New Diagnostics (FIND), who have partnered with the Cepheid Corporation and the University of Medicine and Dentistry of New Jersey.

2.1. Principle of Gene x-pert Test

Cartridge based nucleic acid amplification test (CB-NAAT, Gene X-pert,) is an automated cartridge-based molecular technique which not only detects Mycobacterium Tuberculosis but also rifampicin resistance within two hours. Gene X-pert test is a highly sensitive and specific test for tuberculosis(TB) diagnosis that detects DNA sequences specific for Mycobacterium tuberculosis and rifampicin resistance i.e. rpoB gene. It is also called Gene X-pert because it works as its name.

2.2. Requirements for Gene X-pert Test

1. Kit contains-

- Cartridge
- Dropper
- and reagent

2. Gene X-pert machine

3. Desktop

4. Bar code reader/ bar code scanner

5. Specimen: sputum, CSF, urine, and other body fluids like pleural fluid, synovial fluid, ascitic fluid, and so on

2.3. Procedure of Gene X-pert Test

1. Mix specimen (sputum) and reagent in equal volume.
2. Wait for 15 minutes.
3. Transfer reagent treated specimen into cartridge sample well by using a dropper. (Volume up to mark in dropper)
4. Go to the desktop and click create test.
5. Scan cartridge bar code by the scanner and fill the form mainly patient name and specimen type.
6. Click the Start test button and finally load the cartridge on the holder as shown by default (A1/A2/A3/A4) also notice clicking orange signal.
7. After that push the door and wait for the result (1 hour 51 minutes)

2.4. Result Interpretation of Test

- MTB not detected
- MTB detected with grading either

1. very low
2. low
3. medium
4. or high with

- Rif Resistance NOT DETECTED or
- Rif Resistance DETECTED

2.5. Advantages of Gene X-pert Test

1. Gene X-pert test is highly sensitive comparatively AFB stain and culture of Mycobacterium tuberculosis. (To be AFB positive load should be in sample 10000 bacteria/ml, in culture 10-100 whereas in gene X-pert only 1-10).
2. Very little technical training is required to operate.

3. Minimized bio hazard comparatively AFB stain and culture. (reagent having 10% sodium hypochlorite not only dissolve the specimen but also minimizes contamination)
4. Time is taken to Mycobacterium culture 2-6 weeks and needs 3 weeks extra for drug resistance test that is minimized in this test within hours. We can dispatch the report within 2 half an hour.

2.6. Disadvantages of Gene X-pert Test

1. The shelf life of the cartridge is only 18 months.
2. A very stable electricity supply is required.
3. Calibrations need annually.
4. Test cost is high.

Annex III: Declaration Form

We, the undersigned, hereby declare that this research paper is our original work and has not been presented for an award of a degree in Wolkite University or any other university.

Name of the students:

1. Name: ABATE TILAHUN Signature: _____ Date: 24/08/2023 G.C

2. Name: TRHAS HABTE Signature: _____ Date: 24/08/2023 G.C

3. Name: MARUFA MUSHK Signature: _____ Date: 24/08/2023 G.C

Approval of the advisors

This research paper will be approved by the advisors:

1. Name: Mr. REBIE KEDIR Signature: _____ Date: 24/08/2023 G.C

2. Name: Mr. ASNAKE SEMENEH Signature: _____ Date: 24/08/2023 G.C

Examiner

1. Name: _____Signature: _____Date_____/_____/_____

2. Name: _____ Signature: _____Date_____/____/_____

Name of School head: _____

Signature: _____ Date of submission: 24/08/2023 GC.