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Age-Independent Molecular Resistance Patterns in Tuberculosis Patients: A Cross-Sectional Analytical Study

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Abstract: **Background:** Tuberculosis (TB) remains a major global public health challenge and further complicated by the emergence of drug-resistant strains. Age may influence drug-resistance patterns due to differences in prior treatment exposure among adults, reactivation of latent infection in the elderly, and ongoing transmission dynamics in younger populations. **Objective:** To evaluate age-wise differences in the distribution of resistance-associated genetic markers—*rpoB, katG, inhA, gyrA, gyrB, rrs,* and *eis*—among bacteriologically confirmed tuberculosis patients. **Methods:** A laboratory-based cross-sectional analytical study was conducted among 317 confirmed TB patients referred for molecular drug susceptibility testing between January and December 2024. Patients were stratified into four age groups (≤17, 18–40, 41–60, and ≥61 years). Genomic DNA extracted from clinical specimens was analyzed using GenoType MTBDRplus Version 2.0 and MTBDRsl Version 2.0 line probe assays. Age-wise distributions of wild-type (WT) and mutant (MUT) probes were compared using the Chi-square test. **Results:** Across all age groups, WT probes were predominated for all resistance-associated genes analyzed. No statistically significant association was observed between age and most molecular resistance markers ($p>0.05$). A significant age-wise association was identified only for *rpoB* MUT1 $p = 0.022$, with higher mutation frequency among patients aged 18–40 years and ≥61 years. Mutations associated with isoniazid, fluoroquinolone, and injectable drug resistance did not vary significantly across age categories. **Conclusion:** Age was not a major determinant of molecular drug resistance patterns in tuberculosis patients, except for rpoB MUT1. These findings indicate that molecular drug susceptibility testing should be applied uniformly across all age groups, and diagnostic decisions should not be influenced by patient age alone. Larger, multicentric age-stratified studies are warranted to detect subtle epidemiological trends in drug-resistant TB.

Key Words: Tuberculosis, Age Analysis, Molecular Drug Resistance, Line Probe Assay, *rpoB*, *katG*, *inhA*

INTRODUCTION

Tuberculosis (TB) continues to be one of the leading causes of infectious disease–related morbidity and mortality worldwide, particularly in low- and middle-income countries. The growing burden of drug-resistant TB—including multidrug-resistant (MDR) and extensively drug-resistant (XDR) forms—poses a significant threat to global TB elimination efforts [1–3]. Rapid identification of drug resistance is therefore critical for timely initiation of effective treatment and interruption of transmission.

Resistance to first- and second-line anti-tubercular drugs in *Mycobacterium tuberculosis* is primarily mediated by specific genetic mutations. Rifampicin resistance is associated with mutations in the *rpoB* gene, while resistance to isoniazid involves alterations in *katG* and the *inhA* promoter region. Fluoroquinolone resistance is linked to mutations in *gyrA* and *gyrB*, and resistance to injectable drugs such as aminoglycosides is associated with mutations in *rrs* and *eis* genes [4–10]. Line probe assays (LPAs) exploit these molecular targets and are endorsed by the World Health Organization for routine TB diagnostics.

Age is an important epidemiological factor for many of the viral, bacterial and including TB transmission dynamics, immune response, treatment exposure, and disease reactivation. Pediatric TB often reflects recent transmission, whereas adult TB—particularly in economically productive age groups—may be associated with increased exposure and treatment interruptions. [11–16]. Despite these distinctions, evidence on age-specific molecular resistance patterns remains limited.

The age-specific distribution of resistance-associated mutations might yield some information about the transmission patterns, program gaps, and interventions. Biological issues in older adults (e.g. immune senescence in old age) as well as more prevalent comorbidities (e.g. diabetes and chronic lung disease) and heightened susceptibility to reactivation of latent tuberculosis infection may affect treatment outcome and the acquisition or maintenance of resistant strains. The aim of this study was hence to evaluate the relationship between age and molecular drug resistance markers of tuberculosis patients based on a line probe assay diagnostic.

Aim and Objectives

Aim: To assess age based differences in the presence and absence of resistance associated genetic markers among tuberculosis patients.

Objectives

This study aims to determine the prevalence of *rpoB, katG, inhA, gyrA, gyrB, rrs,* and *eis* gene targets across different age groups, to compare the age-wise distribution of wild-type and mutant probes detected by line probe assay, and to evaluate the statistical association between age and resistance-associated molecular markers.

METHODS

This cross-sectional analytical study was conducted at a State TB Demonstration and Training Centre–Intermediate Reference Laboratory (STDC-IRL), Bangalore, India functioning as a regional diagnostic hub under the National TB Elimination Program.

Study Population

A total of 317 bacteriologically confirmed tuberculosis patients referred for molecular drug resistance testing were included. Both newly diagnosed and previously treated patients across all age groups were included.

Inclusion Criteria

All the patients with microbiologically confirmed *Mycobacterium tuberculosis* infection are included.

Exclusion Criteria

Samples that were contaminated or had incomplete demographic or laboratory data were excluded from the study.

Specimen Collection and Processing

Clinical specimens, predominantly sputum, were collected following national TB program guidelines. Samples were processed using the NALC–NaOH decontamination method, and genomic DNA was extracted as per manufacturer instructions.

Molecular Testing

Drug resistance–associated gene regions were detected using GenoType MTBDRplus and GenoType MTBDRsl assays (Hain Lifescience GmbH, Germany) [17-18]. Both wild-type and mutant probes related to rifampicin, isoniazid, fluoroquinolone, and injectable drug resistance were analyzed. Results were interpreted strictly according to manufacturer and WHO-recommended guidelines.

Outcome Measures

The primary outcome of the study was the presence or absence of resistance-associated genetic markers stratified by age group, while the secondary outcome focused on the age-wise distribution of wild-type (WT) and mutant (MUT) probes detected by the line probe assay.

Data Management and Statistical Analysis

Data were entered into Microsoft Excel and analyzed using SigmaPlot version 14.5 software. Patients were categorized into four age groups: ≤17 years (n = 25), 18–40 years (n = 136), 41–60 years (n = 117), and ≥61 years (n = 39). Categorical variables were expressed as frequencies and percentages. Associations between age and molecular markers were assessed using the Chi-square test. A p-value ≤0.05 or less was considered statistically significant [19-20].

RESULTS

The presence and absence of the *rpoB* gene and its corresponding wild-type (WT) probes among 317 tuberculosis patients is given in Table 1. Overall, the *rpoB* gene was detected in the majority of patients across all age groups, with presence rates of 84.0% (21/25) in those aged ≤17 years, 87.5% (119/136) in the 18–40-year group, 86.3% (101/117) among 41–60-year-olds, and 82.1% (32/39) in patients aged ≥61 years. Statistical analysis revealed no significant association between age and overall *rpoB* gene presence ($p = 0.777$). Similarly, the distribution of individual *rpoB* wild-type probes (WT1 to WT8) showed a consistent predominance of WT probe presence across all age categories, with no statistically significant age-wise differences observed ($p>0.05$ for all comparisons). The highest WT probe positivity was generally observed in the adult age groups (18–40 and 41–60 years), while slightly lower but comparable frequencies were noted in pediatric and elderly patients. Collectively, these findings indicate that the presence of the *rpoB* gene and its wild-type regions does not vary significantly with age, suggesting an age-independent distribution of rifampicin susceptibility–associated molecular targets in the study population.

Distribution of *rpoB* mutant probes among tuberculosis patients given in Table 2. Overall, *rpoB* mutations were infrequently detected across all age groups. A statistically significant association with age was observed only for *rpoB* MUT1 ($p = 0.022$), with mutations identified predominantly in patients aged 18–40 years and ≥61 years, while none were detected in the 41–60-year group. In contrast, no significant age-wise associations were observed for *rpoB* MUT2A, MUT2B, or MUT3 ($p>0.05$), as these mutations were rare and uniformly distributed across age categories. These findings suggest that, except for *rpoB* MUT1, rifampicin

Table 1: The Presence and Absence of Rpob among Tuberculosis Patients, in Various Age Groups

S.No.	Variable	Age group	Present	Absent	Statistical analysis
1	rpoβ	< 17 years	21	4	χ2 = 1.101 p = 0.777
		18–40 years	119	16	
		41–60 years	101	16	
		> 61 years	32	7	
2	rpoβ WT 1	< 17 years	23	2	χ2 = 0.873 p = 0.832
		18–40 years	116	20	
		41–60 years	100	17	
		> 61 years	33	6	
3	rpoβ WT 2	< 17 years	22	3	χ2 = 0.487 p = 0.922
		18–40 years	114	22	
		41–60 years	101	16	
		> 61 years	33	6	
4	rpoβ WT 3	< 17 years	22	3	χ2 = 1.357 p = 0.716
		18–40 years	111	25	
		41–60 years	100	17	
		> 61 years	34	5	
5	rpoβ WT 4	< 17 years	21	4	χ2 = 1.277 p = 0.735
		18–40 years	111	25	
		41–60 years	101	16	
		> 61 years	33	5	
6	rpoβ WT 5	< 17 years	21	4	χ2 = 1.667 p = 0.644
		18–40 years	111	25	
		41–60 years	102	15	
		> 61 years	33	5	
7	rpoβ WT 6	< 17 years	21	4	χ2 = 0.708 p = 0.871
		18–40 years	114	22	
		41–60 years	102	15	
		> 61 years	34	5	
8	rpoβ WT 7	< 17 years	21	4	χ2 = 0.956 p = 0.812
		18–40 years	105	31	
		41–60 years	95	22	
		> 61 years	31	8	
9	rpoβ WT 8	< 17 years	19	5	χ2 = 2.003 p = 0.572
		18–40 years	89	47	
		41–60 years	82	35	
		> 61 years	27	12	

n -≤ 17 years: 25, 18–40 years: 136, 41–60 years: 117, ≥ 61 years: 39

Table 2: The Presence and Absence of Rpob Mutations among Tuberculosis Patients, in Various Age Groups

S.No.	Variable	Age group	Present	Absent	Statistical analysis
1	rpoβ MUT 1	< 17 years	2	23	χ2 = 9.608 p = 0.022
		18–40 years	2	134	
		41–60 years	0	117	
		> 61 years	2	37	
2	rpoβ MUT 2A	< 17 years	0	25	χ2 = 0.523 p = 0.914
		18–40 years	1	135	
		41–60 years	1	116	
		> 61 years	0	39	
3	rpoβ MUT 2B	< 17 years	0	25	χ2 = 1.054 p = 0.788
		18–40 years	2	134	
		41–60 years	1	116	
		> 61 years	1	38	
4	rpoβ MUT 3	< 17 years	3	22	χ2 = 2.318 p = 0.509
		18–40 years	23	113	
		41–60 years	19	98	
		> 61 years	3	36	

n -≤ 17 years: 25, 18–40 years: 136, 41–60 years: 117, ≥ 61 years: 39

resistance–conferring mutations in the *rpoB* gene do not show a strong age-dependent pattern in the study population.

Distribution of *rpoB* mutant probes among tuberculosis patients given in Table 2. Overall, *rpoB* mutations were infrequently detected across all age groups. A statistically significant association with age was observed only for *rpoB* MUT1 ($p = 0.022$), with mutations identified predominantly in patients aged 18–40 years and ≥61 years, while none were detected in the 41–60-year group. In contrast, no significant age-wise associations were observed for *rpoB* MUT2A,

Table 3: The Presence and Absence of Kat G among Tuberculosis Patients, in Various Age Groups

S.No.	Variable	Age group	Present	Absent	Statistical analysis
1	kat G	< 17 years	20	5	χ2 = 0.429 p = 0.934
		18–40 years	110	26	
		41–60 years	98	19	
		> 61 years	32	7	
2	kat G WT	< 17 years	17	8	χ2 = 2.124 p = 0.547
		18–40 years	72	64	
		41–60 years	66	51	
		> 61 years	23	16	
3	kat G MUT 1	< 17 years	4	21	χ2 = 4.406 p = 0.221
		18–40 years	46	89	
		41–60 years	34	83	
		> 61 years	15	24	
4	kat G MUT 2	< 17 years	1	24	χ2 = 3.022 p = 0.388
		18–40 years	6	130	
		41–60 years	2	115	
		> 61 years	0	39	

n -≤ 17 years: 25, 18–40 years: 136, 41–60 years: 117, ≥ 61 years: 39

Table 4: The Presence and Absence of Inh A among Tuberculosis Patients, in Various Age Groups

S.No.	Variable	Age group	Present	Absent	Statistical analysis
1	inh A	< 17 years	22	3	χ2 = 0.429 p = 0.934
		18–40 years	114	21	
		41–60 years	98	19	
		> 61 years	32	7	
2	inh WT 1	< 17 years	18	7	χ2 = 1.096 p = 0.778
		18–40 years	105	31	
		41–60 years	92	25	
		> 61 years	28	11	
3	inh WT 2	< 17 years	21	4	χ2 = 0.826 p = 0.843
		18–40 years	112	24	
		41–60 years	97	20	
		> 61 years	30	9	
4	inh MUT 1	< 17 years	3	22	χ2 = 1.971 p = 0.579
		18–40 years	10	126	
		41–60 years	8	108	
		> 61 years	5	34	
5	inh MUT 2	< 17 years	0	25	NA
		18–40 years	0	136	
		41–60 years	0	116	
		> 61 years	0	39	
6	inh MUT 3A	< 17 years	0	25	NA
		18–40 years	0	136	
		41–60 years	0	116	
		> 61 years	0	39	
7	inh MUT 3B	< 17 years	0	25	χ2 = 2.745 p = 0.433
		18–40 years	1	135	
		41–60 years	3	113	
		> 61 years	0	39	

n -≤ 17 years: 25, 18–40 years: 136, 41–60 years: 117, ≥ 61 years: 39, NA: Not Applicable

MUT2B, or MUT3 (p>0.05), as these mutations were rare and uniformly distributed across age categories. These findings suggest that, except for *rpoB* MUT1, rifampicin resistance–conferring mutations in the *rpoB* gene do not show a strong age-dependent pattern in the study population.

Distribution of the *katG* gene and its corresponding wild-type and mutant probes among tuberculosis patients are given in Table 3. The *katG* gene was detected in the majority of patients across all age groups, with no statistically significant association between age and overall *katG* presence (p = 0.934). Similarly, the distribution of *katG* wild-type (WT) and mutant probes (MUT1 and MUT2) did not differ significantly across age categories (p>0.05 for all comparisons). Although adult patients aged 18–40 and 41–60 years exhibited higher absolute numbers of *katG* mutations, comparable proportional distributions were observed among pediatric and elderly patients. These findings suggest that isoniazid resistance–associated *katG* gene alterations are not significantly influenced by age in the study population.

The age-wise distribution of the *inhA* gene along with its corresponding wild-type and mutant probes among tuberculosis patients given in Table 4. The *inhA* gene was identified in most patients across all age categories, with no

Table 5: The Presence and Absence of Gyr A among Tuberculosis Patients, in Various Age Groups

S.No.	Variable	Age group	Present	Absent	Statistical analysis
1	gyr A	< 17 years	18	7	χ2 = 3.148 p = 0.369
		18–40 years	107	28	
		41–60 years	100	17	
		> 61 years	32	7	
2	gyr A WT 1	< 17 years	19	6	χ2 = 1.057 p = 0.787
		18–40 years	110	25	
		41–60 years	98	19	
		> 61 years	33	6	
3	gyr A WT 2	< 17 years	18	7	χ2 = 2.971 p = 0.396
		18–40 years	101	34	
		41–60 years	97	20	
		> 61 years	30	9	
4	gyr A WT 3	< 17 years	19	6	χ2 = 0.679 p = 0.878
		18–40 years	102	33	
		41–60 years	93	24	
		> 61 years	31	8	
5	gyr A MUT 1	< 17 years	0	25	χ2 = 6.126 p = 0.106
		18–40 years	5	130	
		41–60 years	0	117	
		> 61 years	2	37	
6	gyr A MUT 2	< 17 years	0	25	χ2 = 5.151 p = 0.161
		18–40 years	0	135	
		41–60 years	3	114	
		> 61 years	0	39	
7	gyr A MUT 3 A	< 17 years	0	25	χ2 = 1.396 p = 0.706
		18–40 years	3	132	
		41–60 years	1	116	
		> 61 years	1	38	
8	gyr A MUT 3 B	< 17 years	0	25	χ2 = 2.699 p = 0.440
		18–40 years	2	133	
		41–60 years	0	117	
		> 61 years	0	39	
9	gyr A MUT 3 C	< 17 years	0	25	χ2 = 2.038 p = 0.565
		18–40 years	5	130	
		41–60 years	7	110	
		> 61 years	2	37	
10	gyr A MUT 3 D	< 17 years	0	25	χ2 = 1.345 p = 0.718
		18–40 years	1	134	
		41–60 years	0	117	
		> 61 years	0	39	

n -≤ 17 years: 25, 18–40 years: 136, 41–60 years: 117, ≥ 61 years: 39, NA: 39(Not Applicable)

Table 6: The Presence and Absence of Gyr B among Tuberculosis Patients, in Various Age Groups

S.No.	Variable	Age group	Present	Absent	Statistical analysis
1	Gyr B	< 17 years	21	4	χ2 = 0.807 p = 0.848
		18–40 years	108	27	
		41–60 years	98	19	
		> 61 years	31	8	
2	gyr B WT	< 17 years	20	5	χ2 = 1.164 p = 0.762
		18–40 years	106	29	
		41–60 years	98	19	
		> 61 years	32	7	
3	gyr B MUT 1	< 17 years	0	25	NA
		18–40 years	0	135	
		41–60 years	0	117	
		> 61 years	0	39	
4	gyr B MUT 2	< 17 years	0	25	NA
		18–40 years	0	135	
		41–60 years	0	117	
		> 61 years	0	39	

n -≤ 17 years: 25, 18–40 years: 136, 41–60 years: 117, ≥ 61 years: 39, NA: Not Applicable

statistically significant association between age and overall *inhA* detection (p = 0.934). Similarly, the prevalence of *inhA* wild-type probes (WT1 and WT2) did not vary significantly with age (p>0.05). Mutations within the *inhA* promoter

Table 7: The Presence and Absence of rrs among Tuberculosis Patients, in Various Age Groups

S.No.	Variable	Age group	Present	Absent	Statistical analysis
1	rrs	< 17 years	19	6	χ2 = 1.323 p = 0.724
		18–40 years	111	24	
		41–60 years	99	18	
		> 61 years	31	8	
2	rrs WT 1	< 17 years	21	4	χ2 = 1.655 p = 0.647
		18–40 years	105	30	
		41–60 years	98	19	
		> 61 years	31	8	
3	rrs WT 2	< 17 years	20	5	χ2 = 0.724 p = 0.867
		18–40 years	111	24	
		41–60 years	99	18	
		> 61 years	31	8	
4	rrs MUT 1	< 17 years	0	25	χ2 = 5.547 p = 0.136
		18–40 years	6	129	
		41–60 years	1	116	
		> 61 years	0	39	
5	rrs MUT 2	< 17 years	0	25	NA
		18–40 years	0	135	
		41–60 years	0	117	
		> 61 years	0	39	

n -≤ 17 years: 25, 18–40 years: 136, 41–60 years: 117, ≥ 61 years: 39, NA: Not Applicable

Table 8: The Presence and Absence of Eis Among Tuberculosis Patients, in Various Age Groups.

S.No.	Variable	Age group	Present	Absent	Statistical analysis
1	eis	< 17 years	21	4	χ2 = 0.629 p = 0.890
		18–40 years	111	24	
		41–60 years	99	18	
		> 61 years	31	8	
2	eis WT 1	< 17 years	21	4	χ2 = 0.420 p = 0.936
		18–40 years	111	24	
		41–60 years	98	19	
		> 61 years	31	8	
3	eis WT 2	< 17 years	20	5	χ2 = 0.474 p = 0.925
		18–40 years	111	24	
		41–60 years	98	19	
		> 61 years	31	8	
4	eis WT 3	< 17 years	18	7	χ2 = 2.364 p = 0.500
		18–40 years	110	25	
		41–60 years	99	18	
		> 61 years	31	8	
5	eis MUT 1	< 17 years	0	25	χ2 = 2.699 p = 0.440
		18–40 years	2	133	
		41–60 years	0	117	
		> 61 years	0	39	

n -≤ 17 years: 25, 18–40 years: 136, 41– 60 years: 117, ≥ 61 years: 39

region were uncommon, with *inhA* MUT1 observed at low and comparable frequencies across age groups and showing no significant age-related association (p = 0.579). Other mutant probes, including MUT2, MUT3A, and MUT3B, were either absent or detected at very low frequencies, limiting age-wise analysis. Collectively, these results suggest that *inhA*-mediated isoniazid resistance exhibits an age-independent distribution in the study population.

Distribution of the *gyrA* gene and its corresponding wild-type and mutant probes among tuberculosis patients given in Table 5. The *gyrA* gene was detected in the majority of patients across all age groups, and no statistically significant association was observed between age and overall *gyrA* presence (p = 0.369). Similarly, the distribution of *gyrA* wild-type probes (WT1, WT2, and WT3) showed no significant age-related variation (p>0.05 for all comparisons). Mutations associated with fluoroquinolone resistance were infrequently detected, with *gyrA* MUT1, MUT2, and MUT3 variants occurring at low frequencies and without statistically significant differences across age categories (p>0.05). Overall, these findings indicate that *gyrA*-mediated fluoroquinolone resistance does not exhibit a significant age-dependent pattern in the study population.

Distribution of the *gyrB* gene and its associated wild-type and mutant probes among tuberculosis patients given Table 6. The *gyrB* gene was detected in a high proportion of cases across all age groups, with no statistically significant association between age and overall *gyrB* presence (p = 0.848). Similarly, the distribution of the *gyrB* wild-type probe did not vary significantly across age categories (p = 0.762). Notably, no *gyrB* mutant probes (MUT1 or MUT2) were detected in any age group, precluding statistical comparison. These findings indicate a uniform, age-independent distribution of *gyrB* gene integrity, suggesting minimal contribution of *gyrB*-mediated fluoroquinolone resistance in the study population.

Distribution of the *rrs* gene and its associated wild-type and mutant probes among tuberculosis patients given in Table 7. The *rrs* gene was detected in the majority of patients across all age groups, with no statistically significant association observed between age and overall *rrs* presence ($p = 0.724$). Similarly, the distribution of *rrs* wild-type probes (WT1 and WT2) did not differ significantly across age categories ($p>0.05$). Mutations associated with resistance to injectable drugs were rare, with *rrs* MUT1 detected infrequently and showing no significant age-related association ($p = 0.136$), while *rrs* MUT2 was not detected in any age group. Overall, these findings indicate that *rrs*-mediated resistance patterns are uncommon and do not exhibit a significant age-dependent variation in the study population.

Distribution of the *eis* gene and its associated wild-type and mutant probes among tuberculosis patients given in Table 8. The *eis* gene was detected in most patients across all age groups, with no statistically significant association between age and overall *eis* presence ($p = 0.890$). Similarly, the distribution of *eis* wild-type probes (WT1, WT2, and WT3) showed no significant age-related variation ($p>0.05$ for all comparisons). Mutations in the *eis* gene were rare, with *eis* MUT1 detected only in a small number of patients in the 18–40-year age group and no significant association with age ($p = 0.440$). These findings suggest that *eis*-mediated resistance to injectable drugs does not demonstrate a significant age-dependent pattern in the study population. A total of 317 tuberculosis patients were included in the age-wise molecular analysis. Across all resistance-associated genes studied, wild-type probe patterns predominated irrespective of age group.

DISCUSSION

This study provides a comprehensive age-stratified molecular analysis of resistance-associated genetic markers in tuberculosis patients. The predominance of wild-type probes across all age groups suggests similar baseline susceptibility patterns irrespective of age. The significant association observed only for *rpoB* MUT1 may reflect differential rifampicin exposure or treatment interruption patterns among specific age groups.

Previous studies have reported higher MDR-TB prevalence among young and middle-aged adults, consistent with the higher mutation burden observed in the 18–40-year age group [14,15]. Lower mutation frequencies among paediatric patients likely reflect limited prior drug exposure, while elderly patients may represent reactivation of older, drug-susceptible infections.

This study provides a comprehensive age-stratified molecular assessment of resistance-associated genetic markers for both first- and second-line anti-tubercular drugs using line probe assay. Unlike most previous studies that primarily focus on treatment history or resistance patterns without demographic stratification, the present analysis systematically evaluates age-wise distributions of wild-type and mutant probes across multiple resistance-associated genes (*rpoB, katG, inhA, gyrA, gyrB, rrs,* and *eis*). The identification of a significant age-related association limited to *rpoB* MUT1 highlights a nuanced epidemiological finding that adds new insight into the molecular dynamics of rifampicin resistance across age groups.

Age-related variations in pathogen prevalence and antimicrobial resistance have been documented across a wide range of infectious diseases, providing a relevant comparative perspective for tuberculosis. In bacterial infections, including Mycobacterium tuberculosis, higher antimicrobial resistance rates are commonly observed among young and middle-aged adults, likely due to cumulative drug exposure, sustained transmission, and treatment interruptions. In contrast, paediatric populations generally exhibit lower resistance levels because of limited prior antimicrobial exposure, while elderly individuals may present with reactivation of earlier, drug-susceptible infections.

Parasitic diseases also demonstrate age-dependent patterns, with children often experiencing higher infection burdens due to immature immune responses, whereas resistance trends are more closely linked to repeated mass drug administration in older age groups. Viral infections show distinct age-related dynamics, where children may have higher infection rates but lower resistance owing to limited antiviral exposure, while resistance-associated mutations are more frequently detected in adults with chronic or recurrent infections such as HIV or hepatitis. Similarly, fungal infections tend to show higher prevalence and antifungal resistance among elderly and immunocompromised individuals, with pediatric populations being less commonly affected. Collectively, these observations underscore age as a key epidemiological factor influencing pathogen exposure, immune responses, and the emergence of antimicrobial resistance across diverse infectious agents.

Mycobacterium leprae, the etiological agent of leprosy, exhibits distinct age-related epidemiological and resistance-associated characteristics. Leprosy is most frequently diagnosed in young and middle-aged adults, a pattern that likely reflects the organism's prolonged incubation period and sustained transmission within communities, while the occurrence of paediatric cases indicates on-going transmission in endemic regions. Paediatric patients generally demonstrate lower levels of drug resistance, which may be attributed to limited prior exposure to multidrug therapy, whereas resistance-associated mutations are more commonly detected among adults, particularly in cases involving relapse, treatment default, or repeated therapeutic exposure. In elderly individuals, clinical disease may represent reactivation of infection acquired earlier in life. Collectively, these observations underscore the role of cumulative drug exposure, host immune status, and duration of infection in shaping disease manifestation and the development of antimicrobial resistance in M. leprae, and highlight the importance of age-stratified surveillance for effective leprosy control programs.

Age-related variations in pathogen prevalence and antimicrobial resistance have been documented across a wide range of infectious diseases, providing a relevant comparative perspective for tuberculosis. In bacterial infections, including Mycobacterium tuberculosis, higher antimicrobial resistance rates are commonly observed among young and middle-aged adults, likely due to cumulative drug exposure, sustained transmission, and treatment interruptions, whereas paediatric populations generally exhibit lower resistance levels because of limited prior antimicrobial exposure, and elderly individuals may present with reactivation of earlier, drug-susceptible infections. Parasitic diseases also demonstrate age-dependent patterns, with children often experiencing higher infection burdens due to immature immune responses, whereas resistance trends are more closely linked to repeated mass drug administration in older age groups. Viral infections show distinct age-related dynamics, where children may have higher infection rates but lower resistance owing to limited antiviral exposure, while resistance-associated mutations are more frequently detected in adults with chronic or recurrent infections such as HIV or hepatitis. Similarly, fungal infections tend to show higher prevalence and antifungal resistance among elderly and immunocompromised individuals, with paediatric populations being less commonly affected. Collectively, these observations underscore age as a key epidemiological factor influencing pathogen exposure, immune responses, and the emergence of antimicrobial resistance across diverse infectious agents.

Mycobacterium leprae, the etiological agent of leprosy, exhibits distinct age-related epidemiological and resistance-associated characteristics. Leprosy can occur across all age groups, but cases are frequently reported in young and middle-aged adults, reflecting the organism's prolonged incubation period and sustained community transmission [21,22]. The occurrence of paediatric cases, particularly in children under 15 years of age, indicates ongoing transmission in endemic regions [22-23]. Paediatric patients generally demonstrate lower levels of drug resistance, likely attributable to limited prior exposure to multidrug therapy, whereas resistance-associated mutations are more commonly detected among adults, particularly in cases involving relapse, treatment default, or repeated therapeutic exposure [21,24]. In elderly individuals, clinical disease may represent reactivation of infection acquired earlier in life, with multibacillary forms and older age groups being observed in epidemiological studies [25-26]. Collectively, these observations underscore the role of cumulative drug exposure, host immune status, and duration of infection in shaping disease manifestation and the development of antimicrobial resistance in M. leprae, and highlight the importance of age-stratified surveillance for effective leprosy control programs.

The findings of this study have important programmatic and clinical implications. The largely age-independent distribution of molecular resistance markers supports the uniform application of rapid molecular diagnostics across all age groups without the need for age-specific testing algorithms. Early and accurate detection of drug resistance remains critical for effective TB management, and these results reinforce the reliability of line probe assays for guiding treatment decisions in paediatric, adult, and elderly populations alike. The observed association of rpoB MUT1 with specific age groups underscores the need for continued surveillance of rifampicin resistance patterns, particularly among economically productive and elderly populations who may have higher exposure or treatment-related risk factors.

CONCLUSION

The present study demonstrates that molecular drug-resistance patterns in *Mycobacterium tuberculosis* are largely consistent across different age groups. Resistance-associated genes, including rpoB, katG, inhA, gyrA, gyrB, rrs, and eis, along with their corresponding wild-type probes, predominated among paediatric, adult, and elderly patients, indicating an overall age-independent distribution of susceptibility-associated molecular targets. Mutations conferring resistance to first- and second-line anti-tubercular drugs were generally infrequent and did not exhibit statistically significant age-wise variation for most genetic markers analyzed, in agreement with previously reported molecular epidemiological findings [6,14,27].

A statistically significant age-related association was observed only for rpoB MUT1, suggesting a limited influence of age on rifampicin resistance. This observation may reflect differences in cumulative drug exposure, treatment interruptions, or transmission dynamics, particularly among economically productive and elderly populations, as documented in earlier drug-resistance surveillance studies [2,14,15]. The lack of significant age-wise variation in mutations associated with isoniazid, fluoroquinolone, and injectable drug resistance further supports the notion that molecular resistance profiles are predominantly shaped by factors other than age alone, such as prior treatment history and programmatic adherence [3,6,16].

Taken together, the largely uniform distribution of molecular resistance markers across age strata underscores the broad applicability of line probe assays for rapid drug-resistance detection across all age groups. These findings support the continued use of standardized molecular diagnostic algorithms within national and global tuberculosis control programs [1,11,28]. Nonetheless, the isolated age-specific association observed for rpoB MUT1 highlights the need for ongoing molecular surveillance and larger, multicentric, age-stratified studies to identify subtle epidemiological trends and to strengthen strategies for the prevention and control of drug-resistant tuberculosis [14,27].

Limitations of the Study

Despite its strengths, this study has certain limitations. The relatively smaller sample size in paediatric and elderly age groups may have limited the power to detect subtle age-related differences in rare resistance mutations. Additionally, clinical variables such as treatment history, HIV status, comorbidities, and adherence patterns were not analyzed in conjunction with molecular findings, which may influence resistance profiles.

Ethical Approval

This study was approved by the Institutional Ethics Committee (Ref. No. 008/12/2021/IEC/SMCH). The requirement for individual informed consent was waived due to the use of anonymized laboratory surveillance data.

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Conflicts of Interest

The authors declare that they have no conflicts of interest.

Author Contributions

HJN was responsible for study conception, laboratory oversight, data analysis, manuscript preparation, and contributed to data curation. GB conducted the literature review. VK managed quality control procedures. NK performed sample processing, analysis, and data compilation. All authors reviewed and approved the final version of the manuscript.

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