JHWCR JOURNAL OF HEALTH, WELLNESS AND COMMUNITY RESEARCH 3007-0570

https://doi.org/10.61919/kkm4wc03

Original Article

Antimicrobial Susceptibility Patterns of Streptococcus pyogenes and Non-Groupable Streptococcus Species Isolated from Clinical Specimens in a Tertiary Care Hospital, Lahore: A Cross-Sectional Study

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"Cite this Article" Received: 06 March 2026; **Accepted**: 18 May 2026; **Published**: 02 June 2026

Author Contributions: Concept: AA, TT, AM; Design: AA, TT, AM; Data Collection: HGZ, FR, IA, SI; Analysis: AM, IA; Drafting: AA, TT, AM; Critical Review and Final Approval: All authors. **Ethical Approval**: The Superior University, Lahore, Pakistan. **Informed Consent**: Written informed consent was obtained from all participants. **Conflict of Interest**: The authors declare no conflict of interest. **Funding**: No external funding; **Data Availability**: Available from the corresponding author on reasonable request; **Acknowledgments**: N/A.

ABSTRACT

Background: Streptococcus pyogenes and other Streptococcus species are clinically important pathogens associated with respiratory, skin, soft-tissue, invasive, and toxin-mediated infections. Local antimicrobial susceptibility data are essential for guiding empirical therapy and antimicrobial stewardship, particularly in tertiary-care settings where antibiotic exposure is high. Objective: To evaluate the distribution and antimicrobial susceptibility patterns of S. pyogenes and non-groupable Streptococcus species isolated from clinical specimens in a tertiary-care hospital in Lahore, Pakistan. Methods: This laboratory-based cross-sectional study was conducted in the Department of Microbiology, Ameer-Ud-Din Medical College/Post Graduate Medical Institute, Lahore, from February 2025 to February 2026. Clinical specimens were cultured on 5% sheep blood agar, and Streptococcus isolates were identified using routine microbiological methods. Antimicrobial susceptibility testing was performed by the Kirby–Bauer disk diffusion method according to CLSI criteria. Data were analyzed using frequencies, percentages, and organism-specific denominators. Results: Among 19,500 processed specimens, 165 were Streptococcus-positive, giving a positivity rate of 0.85%. Of these, 14 isolates were S. pyogenes and 151 were non-groupable Streptococcus species. Non-groupable isolates showed high susceptibility to ceftriaxone, teicoplanin, vancomycin, linezolid, penicillin, and ampicillin, whereas S. pyogenes showed highest susceptibility to teicoplanin, vancomycin, and linezolid. Reduced susceptibility was observed for erythromycin and clindamycin, particularly among S. pyogenes isolates. Conclusion: Penicillin and selected β-lactam agents retained substantial in-vitro activity against streptococcal isolates, while macrolide and lincosamide susceptibility was reduced. Routine susceptibility testing and local antibiogram surveillance are necessary to support rational antibiotic selection. Keywords: Streptococcus pyogenes; non-groupable Streptococcus; antimicrobial susceptibility; antibiotic resistance; penicillin; erythromycin; Kirby–Bauer disk diffusion; Lahore.

INTRODUCTION

Streptococcus species are Gram-positive cocci that commonly occur in chains or pairs and include commensal organisms, opportunistic pathogens, and clinically important invasive pathogens. Their classification is traditionally based on hemolytic activity on blood agar and Lancefield grouping according to cell-wall carbohydrate antigens, which remains useful for differentiating clinically relevant streptococcal groups in routine diagnostic microbiology (1,2). Among human streptococcal pathogens,

Streptococcus pyogenes, or Group A Streptococcus, is particularly important because it causes a broad spectrum of disease ranging from pharyngitis, tonsillitis, impetigo, erysipelas, and cellulitis to invasive and toxin-mediated conditions such as necrotizing fasciitis, bacteremia, puerperal sepsis, pneumonia, septic arthritis, scarlet fever, and streptococcal toxic shock syndrome (3). Non-groupable Streptococcus species, although microbiologically heterogeneous, may also be recovered from clinical specimens and can contribute to diagnostic and therapeutic uncertainty, particularly when susceptibility patterns are not routinely documented in local settings.

The clinical significance of S. pyogenes extends beyond acute infection because untreated or inadequately treated disease may lead to serious non-suppurative complications, including acute rheumatic fever, rheumatic heart disease, and post-streptococcal glomerulonephritis. Globally, S. pyogenes remains associated with substantial morbidity and mortality, with severe disease contributing considerably to the infectious disease burden in both community and hospital settings (4,5). Early microbiological diagnosis and appropriate antimicrobial therapy are therefore essential to reduce transmission, prevent complications, and improve patient outcomes. β-lactam antibiotics, particularly penicillin and amoxicillin, have historically remained first-line agents for confirmed S. pyogenes infections, while macrolides, lincosamides, glycopeptides, oxazolidinones, and cephalosporins are used selectively depending on clinical context, allergy status, infection severity, and susceptibility results (5,6).

Antimicrobial resistance among streptococci has become an increasing concern for clinical microbiology and antimicrobial stewardship. Although S. pyogenes has generally retained susceptibility to penicillin in most global reports, reduced susceptibility or resistance to alternative agents, especially macrolides, tetracyclines, fluoroquinolones, and clindamycin, has been reported with marked geographic variability (7–9). This variability is clinically important because empirical antibiotic selection in tertiary-care hospitals is often influenced by local resistance patterns, antibiotic availability, prior antimicrobial exposure, and the burden of severe illness. In settings where antibiotics are frequently used without microbiological confirmation, local surveillance data are essential to distinguish reliable first-line agents from antibiotics with declining activity.

Despite the clinical importance of S. pyogenes and other streptococcal isolates, local data from tertiary-care hospitals in Lahore remain limited, particularly regarding comparative susceptibility patterns of S. pyogenes and non-groupable Streptococcus species isolated from routine clinical specimens. The absence of updated local antibiogram data may contribute to empirical prescribing, delayed optimization of therapy, and unnecessary use of broad-spectrum or reserve antibiotics. A focused assessment of streptococcal isolate distribution and antimicrobial susceptibility patterns can therefore support evidence-based treatment decisions, guide institutional antimicrobial stewardship, and provide baseline data for future surveillance.

The present study was conducted to evaluate the antimicrobial susceptibility patterns of S. pyogenes and non-groupable Streptococcus species isolated from clinical specimens in a tertiary-care hospital in Lahore, Pakistan. The primary objective was to determine the frequency of S. pyogenes and non-groupable Streptococcus isolates among Streptococcus-positive specimens and to describe their susceptibility profiles against routinely tested antimicrobial agents. The study further aimed to compare susceptibility patterns between both isolate groups in order to identify antibiotics retaining useful in-vitro activity and agents showing reduced susceptibility in the local hospital setting.

MATERIALS AND METHODS

This laboratory-based cross-sectional observational study was conducted in the Department of Microbiology, Ameer-Ud-Din Medical College/Post Graduate Medical Institute, Lahore, Pakistan, over a one-year period from February 2025 to February 2026. The study was designed to evaluate the distribution and antimicrobial susceptibility patterns of Streptococcus isolates recovered from routine clinical specimens submitted to the microbiology laboratory from patients of all age groups and both

sexes. The study population consisted of patients with clinically suspected streptococcal infection whose specimens were received from intensive care units, surgical wards, and ear, nose, and throat units during the study period. The analytical unit was the non-duplicate Streptococcus isolate recovered from an eligible clinical specimen.

Clinical specimens, including throat swabs and sputum samples, were collected aseptically according to routine laboratory procedures and transported promptly to the microbiology laboratory in appropriate transport media. Specimens were eligible for inclusion if they were received during the defined study period, were accompanied by relevant demographic or clinical request information, and yielded Streptococcus species on culture. Duplicate specimens from the same patient and infectious episode were excluded to avoid repeated counting of the same isolate. Specimens showing contamination or mixed growth that prevented reliable Streptococcus isolation and susceptibility interpretation were also excluded. Consecutive eligible specimens were included to reduce selection bias and to reflect the routine clinical microbiology workload of the tertiary-care setting.

All specimens were inoculated on 5% sheep blood agar and incubated at 35–37°C for 18–24 hours in 5% CO_2. Suspected Streptococcus colonies were initially assessed by colony morphology, hemolytic pattern on blood agar, Gram staining, and catalase reaction. S. pyogenes was operationally identified as a β-hemolytic, catalase-negative, Gram-positive coccus arranged in chains or pairs and categorized according to the routine diagnostic criteria used in the microbiology laboratory. Non-groupable Streptococcus species were defined as Streptococcus isolates recovered from clinical specimens that did not meet the operational laboratory classification for S. pyogenes and were reported as non-groupable or ungrouped streptococci within the routine laboratory workflow. This terminology was standardized throughout the manuscript as “non-groupable Streptococcus species” to avoid inconsistency with terms such as “ungrouped” or “non-grouped.”

Antimicrobial susceptibility testing was performed using the Kirby–Bauer disk diffusion method on Mueller–Hinton agar according to Clinical and Laboratory Standards Institute interpretive criteria (12). Antibiotics were analyzed only when susceptibility results were available for the relevant isolate group and could be linked to the final organism category. The harmonized comparative antibiotic panel included ampicillin, penicillin, cefixime, ceftriaxone, levofloxacin, teicoplanin, vancomycin, erythromycin, linezolid, clindamycin, and doxycycline. Susceptibility results were interpreted as sensitive, intermediate, or resistant according to the applicable breakpoint criteria. For descriptive reporting, susceptible isolates were summarized as frequencies and percentages using the relevant organism-specific denominator, while intermediate and resistant results were retained separately in the laboratory dataset to avoid misclassification. Because the S. pyogenes subgroup was small, susceptibility percentages were interpreted alongside raw counts in the final results to avoid overstating precision.

Demographic variables included age and sex where available in the laboratory or patient record. Microbiological variables included total number of processed specimens, number of Streptococcus-positive specimens, organism category, specimen type, clinical source unit, and antimicrobial susceptibility category for each tested antibiotic. The primary outcome was antimicrobial susceptibility pattern among S. pyogenes and non-groupable Streptococcus isolates. Secondary outcomes included isolate distribution across organism categories and comparison of susceptibility proportions between S. pyogenes and non-groupable Streptococcus species. To improve internal consistency, all percentages were calculated using explicitly stated denominators, including total processed specimens for overall positivity and Streptococcus-positive isolates for organism-category proportions.

Data were entered and analyzed using SPSS software. Continuous variables were summarized as mean and standard deviation or median and interquartile range depending on distribution, while categorical variables were summarized as frequencies and percentages. Differences in categorical variables between S. pyogenes and non-groupable Streptococcus isolates were assessed using the Chi-square test when expected cell counts were adequate; Fisher’s exact test was used for sparse comparisons, particularly for

antibiotic comparisons involving the S. pyogenes subgroup. A p-value of <0.05 was considered statistically significant. Missing demographic or susceptibility values were not imputed and were reported using available-case denominators for each variable. To minimize analytical bias, duplicate isolates were excluded, organism terminology was standardized before analysis, susceptibility data were checked against laboratory records before tabulation, and all organism-specific percentages were recalculated from raw denominators.

The study used routinely collected microbiology laboratory and patient-record data, and all data were handled in anonymized aggregate form during analysis. Patient identifiers were removed before statistical processing, and results were reported only at group level. Laboratory reproducibility was strengthened through use of standardized specimen processing, blood agar culture, disk diffusion susceptibility testing, CLSI-based interpretation, duplicate-isolate exclusion, and denominator-specific reporting of all microbiological and demographic variables.

RESULTS

During the study period from February 2025 to February 2026, a total of 19,500 clinical specimens were received and processed in the Department of Microbiology. Among these, 165 specimens yielded Streptococcus species, giving an overall Streptococcus positivity rate of 0.85%. Of the 165 Streptococcus-positive isolates, 14 were identified as Streptococcus pyogenes, representing 8.48% of Streptococcus-positive isolates, while 151 were categorized as non-groupable Streptococcus species, representing 91.52%. Because the S. pyogenes subgroup contained only 14 isolates, all susceptibility percentages for this group were interpreted alongside raw counts to avoid overestimation of precision.

Table 1. Distribution of Streptococcus isolates among processed clinical specimens

Category	Frequency, n	Denominator	Percentage (%)
Total clinical specimens processed	19,500	19,500	100.00
Streptococcus-positive specimens	165	19,500	0.85
Streptococcus pyogenes isolates	14	165	8.48
Non-groupable Streptococcus isolates	151	165	91.52

Among Streptococcus-positive cases, females accounted for 99 of 165 cases, corresponding to 60.0%, whereas males accounted for 66 cases, corresponding to 40.0%. Age data were available for 127 cases. Among these, patients older than 61 years formed the largest age group, with 66 cases, representing 52.0% of cases with available age data. Patients aged 26–60 years accounted for 51 cases, or 40.2%, while patients younger than 25 years accounted for 10 cases, or 7.9%. The demographic pattern therefore showed a predominance of Streptococcus-positive isolates among females and older adults; however, these findings should be interpreted descriptively because organism-specific age and sex distributions were not available for subgroup comparison.

Table 2. Demographic distribution of Streptococcus-positive cases

Variable	Category	Frequency, n	Denominator	Percentage (%)
Sex	Female	99	165	60.0
Sex	Male	66	165	40.0
Age group	<25 years	10	127	7.9
Age group	26–60 years	51	127	40.2
Age group	>61 years	66	127	52.0

The comparative antimicrobial susceptibility analysis showed that non-groupable Streptococcus species had higher susceptibility than S. pyogenes for most β-lactams and reserve agents. Among S. pyogenes isolates, the highest susceptibility was observed for teicoplanin, vancomycin, and linezolid, each showing susceptibility in 13 of 14 isolates, corresponding to 92.86%. Penicillin, ampicillin, and ceftriaxone each showed susceptibility in 12 of 14 S. pyogenes isolates, corresponding to 85.71%. Moderate susceptibility was observed for cefixime, levofloxacin, and doxycycline, while erythromycin and clindamycin showed lower susceptibility at 35.71% and 42.86%, respectively.

Among non-groupable Streptococcus isolates, ceftriaxone demonstrated susceptibility in all 151 isolates, corresponding to 100.0%. Teicoplanin and vancomycin were each active against 150 of 151 isolates, corresponding to 99.34%, while linezolid was active against 149 of 151 isolates, corresponding to 98.68%. Penicillin and ampicillin also showed high activity, with susceptibility rates of 96.69% and 96.03%, respectively. Reduced susceptibility was observed for erythromycin, with only 75 of 151 isolates susceptible, corresponding to 49.67%, and for clindamycin, with 96 of 151 isolates susceptible, corresponding to 63.58%.

Table 3. Comparative antimicrobial susceptibility pattern of S. pyogenes and non-groupable Streptococcus isolates

Antibiotic	S. pyogenes susceptible, n/N (%)	Non-groupable Streptococcus susceptible, n/N (%)	Risk difference, percentage points (95% CI)	p-value
Ampicillin	12/14 (85.71)	145/151 (96.03)	-10.3 (-36.1 to 0.9)	0.139
Penicillin	12/14 (85.71)	146/151 (96.69)	-11.0 (-36.7 to 0.1)	0.110
Cefixime	11/14 (78.57)	137/151 (90.73)	-12.2 (-38.6 to 2.8)	0.161
Ceftriaxone	12/14 (85.71)	151/151 (100.00)	-14.3 (-39.9 to -3.7)	0.007
Levofloxacin	10/14 (71.43)	103/151 (68.21)	3.2 (-23.8 to 21.8)	1.000
Teicoplanin	13/14 (92.86)	150/151 (99.34)	-6.5 (-30.8 to 0.1)	0.163
Vancomycin	13/14 (92.86)	150/151 (99.34)	-6.5 (-30.8 to 0.1)	0.163
Erythromycin	5/14 (35.71)	75/151 (49.67)	-14.0 (-34.9 to 12.8)	0.406
Linezolid	13/14 (92.86)	149/151 (98.68)	-5.8 (-30.2 to 1.0)	0.235
Clindamycin	6/14 (42.86)	96/151 (63.58)	-20.7 (-43.4 to 5.1)	0.154
Doxycycline	10/14 (71.43)	137/151 (90.73)	-19.3 (-45.6 to -1.5)	0.050

Note: p-values were calculated using Fisher's exact test because of the small S. pyogenes subgroup and sparse cell counts. Risk difference represents the susceptibility percentage in S. pyogenes minus susceptibility percentage in non-groupable Streptococcus species. Negative values indicate lower susceptibility among S. pyogenes isolates.

The most clinically relevant susceptibility difference was observed for ceftriaxone, where all non-groupable Streptococcus isolates were susceptible compared with 12 of 14 S. pyogenes isolates, producing a risk difference of -14.3 percentage points and a statistically significant p-value of 0.007. Doxycycline also showed a lower susceptibility proportion among S. pyogenes isolates than among non-groupable Streptococcus isolates, with susceptibility rates of 71.43% versus 90.73%, respectively, and a risk difference of -19.3 percentage points. This comparison reached borderline statistical significance with a p-value of 0.050. For penicillin and ampicillin, S. pyogenes showed lower susceptibility than non-groupable Streptococcus species by approximately 10–11 percentage points, but these differences were not statistically significant. However, because the S. pyogenes denominator was small, these findings should be interpreted cautiously and verified through confirmatory susceptibility testing where clinically indicated.

Several additional antibiotics showed markedly low susceptibility in one organism category but could not be compared statistically because complete paired susceptibility data were not available for both groups. Among S. pyogenes isolates, co-trimoxazole, imipenem, ceftaroline, and chloramphenicol each showed susceptibility in only 1 of 14 isolates, corresponding to 7.14%. Among non-groupable Streptococcus isolates, fosfomycin showed susceptibility in 15 of 151 isolates, corresponding to 9.93%, while nitrofurantoin showed susceptibility in 20 of 151 isolates, corresponding to 13.25%. These findings indicate poor in-vitro activity of these agents against the tested isolates and suggest that they should not be relied upon for empirical treatment without organism-specific susceptibility confirmation.

Table 4. Additional antibiotics with low reported susceptibility

Antibiotic	Organism group	Susceptible, n/N (%)	Comparative p-value
Co-trimoxazole	S. pyogenes	1/14 (7.14)	Not applicable
Imipenem	S. pyogenes	1/14 (7.14)	Not applicable
Ceftaroline	S. pyogenes	1/14 (7.14)	Not applicable
Chloramphenicol	S. pyogenes	1/14 (7.14)	Not applicable
Fosfomycin	Non-groupable Streptococcus	15/151 (9.93)	Not applicable
Nitrofurantoin	Non-groupable Streptococcus	20/151 (13.25)	Not applicable

Overall, the susceptibility profile demonstrated that penicillin, ampicillin, ceftriaxone, teicoplanin, vancomycin, and linezolid retained high in-vitro activity against most Streptococcus isolates in this setting. Non-groupable Streptococcus species showed consistently high susceptibility to ceftriaxone, teicoplanin, vancomycin, linezolid, penicillin, and ampicillin, all exceeding 96%. In contrast, S. pyogenes isolates showed comparatively lower susceptibility across several agents, particularly erythromycin, clindamycin, doxycycline, cefixime, and the additional low-activity antibiotics. The reduced susceptibility to erythromycin in both groups, especially 35.71% among S. pyogenes and 49.67% among non-groupable Streptococcus species, indicates that macrolides may be unreliable for empirical use in this hospital setting unless supported by susceptibility testing.

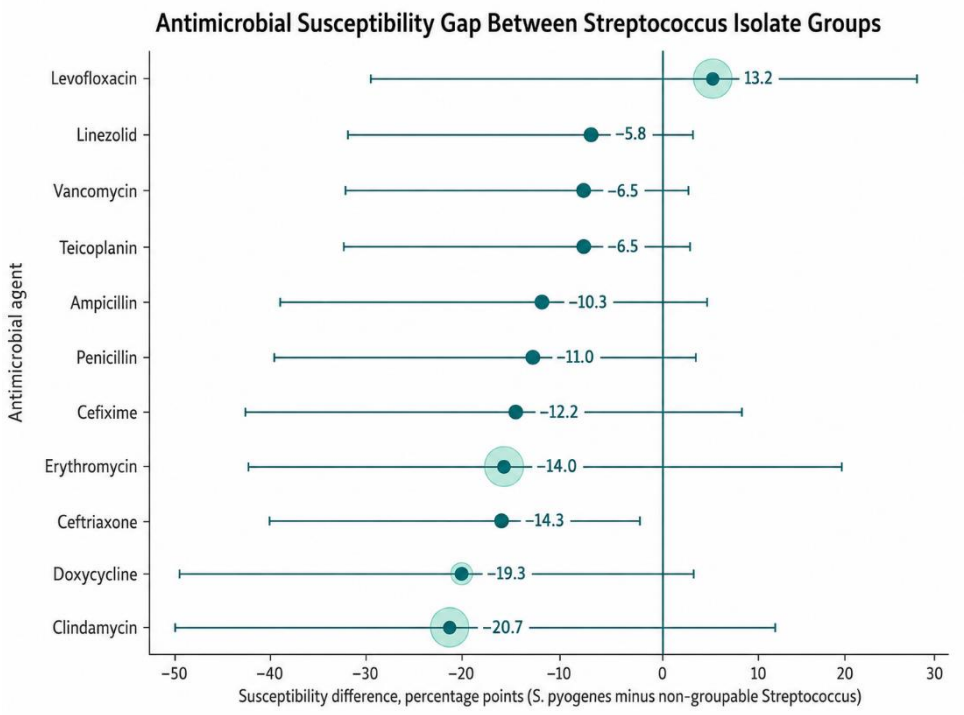

Figure 1. Antimicrobial susceptibility gap between S. pyogenes and non-groupable Streptococcus isolates.

The figure shows lower susceptibility of S. pyogenes for most tested antibiotics, with the largest negative gaps observed for clindamycin, doxycycline, ceftriaxone, erythromycin, cefixime, penicillin, and ampicillin. Levofloxacin showed a small positive gap, while the wide intervals reflect the small S. pyogenes sample size and support cautious interpretation.

Taken together, the results show that Streptococcus species represented a small proportion of total processed clinical specimens but demonstrated clinically meaningful variation in antimicrobial susceptibility by organism category. The strongest retained activity was observed for β-lactams and selected reserve agents, whereas erythromycin, clindamycin, co-trimoxazole, fosfomycin, nitrofurantoin, imipenem, ceftaroline, and chloramphenicol showed limited or inconsistent activity. These findings support routine culture-based susceptibility testing and local antibiogram surveillance to guide rational antimicrobial selection.

DISCUSSION

The present laboratory-based cross-sectional study evaluated the distribution and antimicrobial susceptibility patterns of Streptococcus pyogenes and non-groupable Streptococcus species isolated from clinical specimens in a tertiary-care hospital in Lahore. Among 19,500 processed clinical specimens, 165 yielded Streptococcus species, giving an overall positivity rate of 0.85%. Most Streptococcus-positive isolates were categorized as non-groupable Streptococcus species, while S. pyogenes accounted for 14 isolates, representing 8.48% of Streptococcus-positive isolates. This distribution suggests that S. pyogenes

constituted a relatively small proportion of streptococcal isolates in the submitted clinical specimens, although interpretation should consider the specimen mix, ward-based sampling pattern, and laboratory identification workflow. Because the S. pyogenes subgroup contained only 14 isolates, the susceptibility findings for this organism should be interpreted with caution and preferably presented with raw counts in addition to percentages.

The demographic findings showed a higher frequency of Streptococcus-positive isolates among females and older adults. Females accounted for 99 of 165 cases, while males accounted for 66 cases. Age data were available for 127 cases, among whom patients older than 61 years represented the largest group. This pattern may reflect greater healthcare utilization, comorbidity burden, immune senescence, or hospital exposure among older adults, but these explanations remain inferential because comorbidities, prior antibiotic use, hospitalization duration, and infection severity were not measured in the present dataset. Previous surveillance of severe S. pyogenes disease has shown that older populations may be vulnerable to clinically significant streptococcal infection, particularly in the presence of underlying disease, but the current findings should be interpreted as descriptive isolate-level observations rather than evidence of age-related causality (12).

Penicillin and ampicillin retained high in-vitro activity against the overall streptococcal isolate population, particularly among non-groupable Streptococcus species, where susceptibility exceeded 96%. Among S. pyogenes isolates, penicillin and ampicillin susceptibility was 85.71%, corresponding to 12 of 14 isolates. This finding is clinically important and requires careful interpretation because S. pyogenes has historically remained highly susceptible to penicillin in most global reports, and β-lactams remain central to recommended therapy for confirmed Group A streptococcal infection (5–7). The lower susceptibility proportion observed in this small subgroup may reflect local testing variation, interpretive issues, isolate misclassification, or true reduced susceptibility, and therefore should not be generalized as confirmed penicillin resistance without additional verification. Future work should include confirmatory identification, minimum inhibitory concentration testing, quality-control documentation, and, where feasible, molecular assessment of resistance-associated mechanisms.

Ceftriaxone demonstrated excellent activity among non-groupable Streptococcus species, with susceptibility in all 151 isolates, while 12 of 14 S. pyogenes isolates were susceptible. Glycopeptides and oxazolidinones also showed high in-vitro activity, with teicoplanin, vancomycin, and linezolid demonstrating susceptibility above 92% in S. pyogenes and above 98% in non-groupable Streptococcus species. These findings indicate retained laboratory activity of selected broad-spectrum and reserve agents; however, clinical interpretation must remain stewardship-oriented. Vancomycin, teicoplanin, and linezolid should not be framed as routine empirical options for uncomplicated streptococcal infections, particularly when narrow-spectrum β-lactams remain appropriate for confirmed susceptible infections. Their role should be reserved for selected severe infections, β-lactam contraindication, polymicrobial clinical contexts, or cases guided by susceptibility results and institutional treatment protocols.

Reduced susceptibility to erythromycin was one of the most clinically relevant findings. Only 5 of 14 S. pyogenes isolates and 75 of 151 non-groupable Streptococcus isolates were susceptible to erythromycin, corresponding to susceptibility rates of 35.71% and 49.67%, respectively. This pattern suggests that empirical macrolide use may be unreliable in this hospital setting unless supported by susceptibility testing. Increasing macrolide resistance among streptococci has been reported across multiple regions, with rates varying by geography, population, antimicrobial exposure, and resistance mechanisms (15,16). The current findings are therefore consistent with the broader concern that macrolides should be used cautiously for suspected streptococcal infection, especially in patients labeled as penicillin-allergic, where susceptibility confirmation becomes particularly important.

Clindamycin also showed reduced activity, especially among S. pyogenes isolates, where only 6 of 14 isolates were susceptible. This finding is clinically relevant because clindamycin may be used as an adjunctive agent in severe toxin-mediated Group A streptococcal disease and as an alternative in selected

β-lactam-allergic patients. However, low susceptibility in the current dataset suggests that empirical reliance on clindamycin may be inappropriate without local susceptibility confirmation. Doxycycline also showed lower susceptibility among S. pyogenes than among non-groupable Streptococcus species, while levofloxacin demonstrated only moderate activity in both groups. Similar variability in susceptibility to non-β-lactam agents has been reported in regional and international studies, supporting the need for local antibiogram-based prescribing rather than extrapolation from global averages (8,17–21).

Several agents showed very low reported susceptibility in the available dataset, including co-trimoxazole, imipenem, ceftaroline, and chloramphenicol among S. pyogenes isolates, and fosfomycin and nitrofurantoin among non-groupable Streptococcus species. These results should be interpreted carefully because some of these antibiotics are not standard first-line agents for routine streptococcal infections, and the clinical relevance of testing may vary by specimen type, infection site, and breakpoint availability. The unexpectedly low susceptibility reported for certain agents, particularly imipenem and ceftaroline in S. pyogenes, should prompt verification of organism identification, disk potency, testing conditions, breakpoint selection, and data transcription before firm conclusions are drawn. Rather than labeling these findings as definitive multidrug resistance, the present study more appropriately demonstrates reduced in-vitro susceptibility to selected alternative agents within the tested isolate set.

The main strength of this study is that it provides local tertiary-care laboratory data from a large specimen-processing denominator, offering useful baseline information for antimicrobial stewardship and future surveillance. The study also compares S. pyogenes with non-groupable Streptococcus species, which may help clinicians and microbiologists recognize organism-category variation in susceptibility patterns. Nevertheless, several limitations must be acknowledged. The study was conducted at a single center, which limits generalizability to other hospitals or community settings. The S. pyogenes subgroup was small, making percentage estimates unstable and producing wide uncertainty around comparative estimates. The study did not include molecular identification, resistance-gene detection, minimum inhibitory concentration testing, or detailed clinical correlation with infection severity, prior antibiotic exposure, comorbidities, and treatment outcomes. Age data were incomplete, and specimen-source distribution was not sufficiently detailed to distinguish colonization from clinically confirmed infection in all cases. These limitations should guide cautious interpretation and support the need for multicenter surveillance using standardized organism identification, full susceptibility panels, and quality-controlled antimicrobial testing.

Overall, the findings support continued use of culture-based diagnosis and routine susceptibility testing for clinically suspected streptococcal infections in tertiary-care settings. Penicillin and selected β-lactam agents retained substantial in-vitro activity, particularly among non-groupable Streptococcus species, while erythromycin, clindamycin, and several alternative agents showed reduced susceptibility and should not be used empirically without local evidence or isolate-specific susceptibility confirmation. Future studies should include larger numbers of S. pyogenes isolates, standardized Lancefield grouping or molecular confirmation, MIC-based validation of unexpected susceptibility patterns, and linkage of microbiological results with clinical outcomes to strengthen therapeutic interpretation.

CONCLUSION

This study demonstrated that Streptococcus species accounted for 165 of 19,500 processed clinical specimens in a tertiary-care hospital in Lahore, with S. pyogenes representing 14 isolates and non-groupable Streptococcus species representing 151 isolates. Penicillin, ampicillin, ceftriaxone, teicoplanin, vancomycin, and linezolid showed high in-vitro activity against most streptococcal isolates, although the lower penicillin susceptibility observed among the small S. pyogenes subgroup should be verified through confirmatory testing before being interpreted as evidence of true resistance. Erythromycin, clindamycin, doxycycline, and several additional alternative agents showed reduced or

inconsistent susceptibility, indicating that empirical use of these antibiotics may be unreliable without culture and sensitivity confirmation. The findings emphasize the importance of routine antimicrobial susceptibility testing, local antibiogram surveillance, and antimicrobial stewardship to preserve the effectiveness of narrow-spectrum agents and reduce unnecessary use of broad-spectrum or reserve antibiotics.

REFERENCES

1. Haenni M, Lupo A, Madec JY. Antimicrobial resistance in Streptococcus spp. Microbiol Spectr. 2018;6(2):10.1128/microbiolspec.ARBA-0008-2017.
2. Lancefield RC, Freimer EH. Type-specific polysaccharide antigens of group B streptococci. Epidemiol Infect. 1966;64(2):191-203.
3. Lannes-Costa PS, de Oliveira JS, Santos GS, Nagao PE. A current review of pathogenicity determinants of Streptococcus sp. J Appl Microbiol. 2021;131(4):1600-20.
4. Cunningham MW. Pathogenesis of group A streptococcal infections. Clin Microbiol Rev. 2000;13(3):470-511.
5. World Health Organization. The current evidence for the burden of group A streptococcal diseases. Geneva: World Health Organization; 2005.
6. Carapetis JR, Steer AC, Mulholland EK, Weber M. The global burden of group A streptococcal diseases. Lancet Infect Dis. 2005;5(11):685-94.
7. Siemens N, Lütticken R. Streptococcus pyogenes, a highly adapted human pathogen: potential implications of its virulence regulation for epidemiology and disease management. Pathogens. 2021;10(6):776.
8. Tatara K, Gotoh K, Okumiya K, Teramachi M, Ishimoto K, Tanaka Y, et al. Molecular epidemiology, antimicrobial susceptibility, and characterization of fluoroquinolone non-susceptible Streptococcus pyogenes in Japan. J Infect Chemother. 2020;26(3):280-4.
9. Beres SB, Zhu L, Pruitt L, Olsen RJ, Faili A, Kayal S, et al. Integrative reverse genetic analysis identifies polymorphisms contributing to decreased antimicrobial agent susceptibility in Streptococcus pyogenes. mBio. 2022;13(1):e03618-21.
10. Barros RR. Antimicrobial resistance among beta-hemolytic Streptococcus in Brazil: an overview. Antibiotics. 2021;10(8):973.
11. Clinical and Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing. CLSI supplement M100. Wayne, PA: Clinical and Laboratory Standards Institute.
12. Lamagni TL, Darenberg J, Luca-Harari B, Siljander T, Efstratiou A, Henriques-Normark B, et al. Epidemiology of severe Streptococcus pyogenes disease in Europe. J Clin Microbiol. 2008;46(7):2359-67.
13. Kebede D, Admas A, Mekonnen D. Prevalence and antibiotics susceptibility profiles of Streptococcus pyogenes among pediatric patients with acute pharyngitis at Felege Hiwot Comprehensive Specialized Hospital, Northwest Ethiopia. BMC Microbiol. 2021;21(1):135.
14. Ahmed OB, Asghar AH, Elhassan MM. Epidemiological aspects and antibiotics susceptibility patterns of Streptococcus pyogenes isolated from subjects with tonsillitis, Sudan. Sudan J Med Sci. 2023;18(2):210-20.

15. Arvand M, Hoeck M, Hahn H, Wagner J. Antimicrobial resistance in Streptococcus pyogenes isolates in Berlin. J Antimicrob Chemother. 2000;46(4):621-4.

16. Tadesse M, Hailu Y, Biset S, Ferede G, Gelaw B. Prevalence, antibiotic susceptibility profile and associated factors of group A streptococcal pharyngitis among pediatric patients with acute pharyngitis in Gondar, Northwest Ethiopia. Infect Drug Resist. 2023;16:1637-48.

17. Hussein EF, Raheem HQ, Merdas AH. Antimicrobial susceptibility pattern of isolates of Streptococcus pyogenes obtained from patients with throat infection. J Commun Dis. 2024;56(3):3-9.

18. Camara M, Dieng A, Boye CS. Antibiotic susceptibility of Streptococcus pyogenes isolated from respiratory tract infections in Dakar, Senegal. Microbiol Insights. 2013;6:MBI.S12996.

19. Gebre AB, Fenta DA, Negash AA, Hayile BJ. Prevalence, antibiotic susceptibility pattern and associated factors of Streptococcus pyogenes among pediatric patients with acute pharyngitis in Sidama, Southern Ethiopia. Int J Microbiol. 2024;2024:9282571.

20. Mekuria S, Getu F, Walle M, Tesfaye A, Tolossa D, Abebe T, et al. Throat swab culture positivity rate, antibiotic susceptibility profile, and associated risk factors of Streptococcus pyogenes among children with acute pharyngitis attending Jigjiga University Sheik Hassan Yebere Referral Hospital, Jigjiga, Ethiopia. Int J Microbiol. 2026;2026:8337012.

21. Meles HN, Aregawi BB, Gebregergis MW, Hailekiros H, Weldu Y, Thangaraju P, et al. Prevalence, antimicrobial susceptibility patterns and associated factors of Streptococcus pyogenes among apparently healthy school children in Mekelle city primary schools, Northern Ethiopia. Heliyon. 2024;10(14):e34215.