

## A Study on Isolation of Group A Beta Hemolytic Streptococci and its Sensitivity to Co-Amoxiclav in Acute Tonsillitis

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### ABSTRACT

**Introduction:** Acute tonsillitis, often caused by Group A Beta-Hemolytic Streptococci (GABHS), is a significant global health concern. Co-Amoxiclav is widely used for treatment, but rising antibiotic resistance necessitates evaluation of its efficacy. This study aimed to isolate GABHS and assess its sensitivity to Co-Amoxiclav in patients with acute tonsillitis.

**Methods:** A hospital-based cross-sectional study was conducted at Manipal Teaching Hospital, Nepal, from November 2023 to October 2024 after obtaining ethical clearance (MCOMS/IRC/574/GA). Throat swabs from 280 clinically diagnosed patients with acute tonsillitis were collected and analyzed for bacterial pathogens. Antimicrobial susceptibility testing was performed, and data were analyzed using SPSS.

**Results:** Out of 280 clinically diagnosed patients with acute tonsillitis, GABHS was isolated in 12.85% of cases, with 77.77% sensitivity to Co-Amoxiclav. The chi-square test (p-value = 0.80) indicated no significant difference, suggesting Co-Amoxiclav's effectiveness regardless of GABHS presence in bacterial tonsillitis. Azithromycin showed the highest resistance, while vancomycin was the most effective.

**Conclusions:** GABHS remains a key pathogen in acute tonsillitis, and Co-Amoxiclav is still effective for treatment. However, declining sensitivity underscores the need for judicious antibiotic use and exploration of alternative treatments.

**Keywords:** *Azithromycin; Co-Amoxiclav; Group A beta-hemolytic streptococcus; Sensitivity; Tonsillitis.*



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## INTRODUCTION

Acute tonsillitis, a common condition worldwide, is frequently caused by bacterial infections, particularly Group A Beta-Hemolytic Streptococci (GABHS) also known as *Streptococcus pyogenes*. [1] GABHS infections account for millions of cases annually and can lead to severe complications, such as rheumatic fever and post-streptococcal glomerulonephritis, if untreated. [2] Definitive identification of the causative pathogens is essential for determining the most appropriate treatment plan, especially when antibiotics are involved. [3,4] Accurate pathogen recognition helps avoid unnecessary antibiotic use, which is vital in an era of rising antibiotic resistance.

Penicillin has traditionally been the first-line treatment for GABHS infections, but Co-Amoxiclav, combination of amoxicillin (an amino penicillin having wider coverage than penicillin due its activity against gram negative cocci and enterobacteria) and Clavulanic acid (irreversible suicide inhibitor of beta -lactamase) a broad-spectrum antibiotic, is increasingly prescribed. [5,6] However, the rise in antibiotic resistance necessitates ongoing evaluation of GABHS susceptibility to Co-Amoxiclav.

This study aimed to isolate GABHS from patients with acute tonsillitis and assess its sensitivity to Co-Amoxiclav, providing insights into current antimicrobial resistance trends and guiding treatment decisions.

## METHODS

This hospital-based, prospective, observational, cross-sectional study was conducted at Manipal Teaching Hospital, Pokhara, Nepal, from November 2023 to October 2024. Ethical approval was obtained from the Institutional Review Committee (MCOMS/IRC/574/GA).

Throat swabs were collected from 280 clinically diagnosed cases of acute tonsillitis using sterile techniques. Patients already on Co-Amoxiclav or amoxicillin, those under 5 years of age, fungal cultures, peri tonsillitis and peritonsillar abscess were excluded.

Data were collected from both outpatient and inpatient department using structured proforma specifically designed for the study. The proforma included details such as age, gender, symptoms, type of tonsillitis, lymph nodes examination findings, throat swab results and swab culture reports.

For throat swab collection, patients were asked to sit facing a light source, then tongue was depressed using a tongue depressor, and two sterile cotton-wool swabs were rubbed over each tonsil separately, ensuring that the tongue or buccal surfaces were not touched. Each sample was labeled with the patient's hospital number, name, age, and gender, then transported to the microbiology laboratory under sterile conditions and processed within 2 hours of collection. Swab results were traced by patients during follow-up

visits after 72 hours. For lost follow-up cases, researchers retrieved swab results from the microbiology laboratory and entered them into the proforma sheet. Data were entered into Microsoft Excel 2016 and analyzed using SPSS Statistics for Windows, version 26.0.

## RESULTS

Out of 280 patients with acute tonsillitis, the age ranged from 6 to 67 years, with a mean age of  $26.51 \pm 11.3$  years. Female patients were predominant, with a female-to-male ratio of 149:131. The majority of patients were in the 21-30 years age group (41.07%), followed by 11-20 years age group (21.07%), 31-40 years age group (18.21%), 41-50 years age group (9.29%), less than 10 years (7.86%) and more than 51 years (2.50%). Most patients experienced throat discomfort and fever, other symptoms like neck swelling, voice changes and constitutional symptoms were also present. Most present with multiple complains underscoring the complexity of acute tonsillitis presentations. Enlarged and tender jugulodigastric lymph nodes were observed in 175(62.5%) patients on that 121 were having unilateral rest 54 patients bilateral involvement was seen.

**Table 1. Distribution of Most Common Tonsillitis Symptoms in the Study Population (n = 280)**

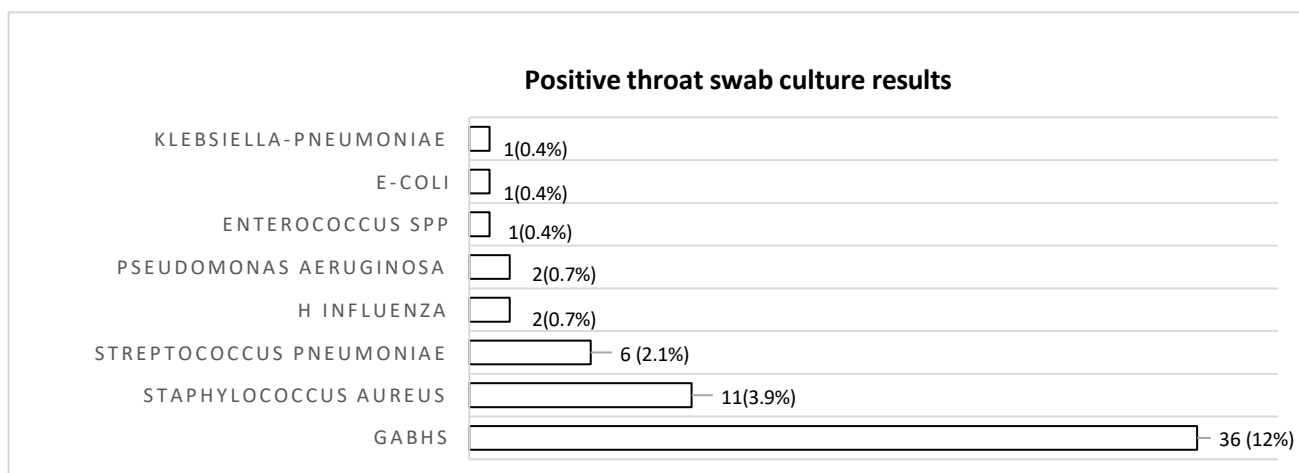
Symptoms	Frequency (n)	Percentage (%)
Throat discomfort	260	92.86
Fever	172	61.43
Jugulodigastric	175	62.50
Lymphadenopathy		
Systemic symptoms*	88	31.43
Localized symptoms**	102	36.43

\*Systemic symptoms: Constitutional symptoms (e.g., fatigue, malaise).

\*\*Localized symptoms: Neck swelling, ear pain, dental pain, voice change.

On clinical examination, the types of acute tonsillitis identified were acute superficial tonsillitis presents as part of generalized pharyngitis (50.71%, n = 142), acute follicular tonsillitis present as yellowish white dots on tonsil (17.50%, n = 49), acute membranous tonsillitis presents as whitish membrane over tonsil (17.14%, n = 48), and acute parenchymal tonsillitis congested enlarged tonsil only (14.64%, n = 41). Throat swab cultures were positive in 60 cases (21.43%), while the remaining 220 cases (78.57%) showed no growth or normal flora. Among the positive cultures, *Streptococcus pyogenes* (GABHS) was isolated in 36 cases (12.86%), followed by *Staphylococcus aureus* (3.93%, n = 11), *Streptococcus pneumoniae* (2.14%, n = 6), *Haemophilus influenzae* (0.71%, n = 2), *Pseudomonas aeruginosa* (0.71%, n = 2), *Enterococcus spp.* (0.36%, n = 1), *Escherichia coli* (0.36%, n = 1), and *Klebsiella pneumoniae* (0.36%, n = 1). Figure 1

**Figure 1: Positive throat swab culture results (n = 60)**



Throat swab positivity rates according to the types of tonsillitis were as follows: acute membranous tonsillitis (10.35%, n = 29), acute follicular tonsillitis (7.50%, n = 21), acute superficial tonsillitis (2.14%, n = 6), and acute parenchymal tonsillitis (1.42%, n = 4) and their specification for GABHS are illustrated on Table 2

**Table 2. Positive bacterial findings(n=60) and GABHS isolation(n=36)**

Types of Tonsillitis	Swab Positive Results	Isolated GABHS	
		Frequency	Percentage
Acute Superficial Tonsillitis	6	2	0.71
Acute Parenchymatous Tonsillitis	4	2	0.71
Acute Follicular Tonsillitis	21	13	4.64
Acute Membranous Tonsillitis	29	19	6.78

Out of the 60-throat swab-positive samples, the microbiology department tested 17 different antibiotics, with 7 antibiotics applied to all cases. The antibiotic sensitivity results for overall swab positive tonsillitis cases were as follows: Azithromycin (50%), Chloramphenicol (73.33%), Co-Amoxiclav (76.66%), Clindamycin (83.33%), Gentamicin (91.66%), levofloxacin (93.33%), and Vancomycin (96.66%). For GABHS isolates specifically, the sensitivity rates were: Azithromycin (50%), Chloramphenicol (77.77%), Co-Amoxiclav (77.77%), Clindamycin (80.55%), Gentamicin (100%), levofloxacin (94.44%), and Vancomycin (97.22%).

**Table 3: Antibiotic Resistance and Sensitivity Profiles in Tonsillitis and GABHS Infections (n = 60 for Tonsillitis; n = 36 for GABHS)**

Antibiotic	Resistant in Tonsillitis (n=60)	Resistant in GABHS (n=36)	Sensitive in Tonsillitis (n=60)	Sensitive in GABHS (n=36)
Azithromycin	30 (50%)	18 (50%)	30 (50%)	18 (50%)

Chloramphenicol	16 (26.66%)	8 (22.22%)	44 (73.33%)	28 (77.77%)
Co-Amoxiclav	14 (23.33%)	8 (22.22%)	46 (76.66%)	28 (77.77%)
Clindamycin	10 (16.66%)	7 (19.44%)	50 (83.33%)	29 (80.55%)
Gentamycin	5 (8.33%)	0 (0%)	55 (91.66%)	36 (100%)

Resistance and sensitivity rates for Levofloxacin and Vancomycin were as follows: Levofloxacin (Resistant: Tonsillitis 4 [6.66%], GABHS 2 [5.55%]; Sensitive: Tonsillitis 56 [93.33%], GABHS 34 [94.44%]); Vancomycin (Resistant: Tonsillitis 2 [3.33%], GABHS 1 [2.77%]; Sensitive: Tonsillitis 58 [96.66%], GABHS 35 [97.22%]).

Using Chi-square test comparing Co-Amoxiclav sensitivity and resistance among swab positive cases of GABHS and other pathogens yielded a p-value of 0.8. This suggests that Co-Amoxiclav remains effective in treating tonsillitis, regardless of the causative organism. (Table 4)

**Table 4: Chi-Square Test Comparing Co-Amoxiclav Sensitivity and Resistance in Throat Swab-Positive Cases of GABHS and Other Pathogens**

Swab Results	Co-Amoxiclav		Total	p-value
	Sensitive	Resistance		
GABHS	28	8	36	0.80
Other Pathogens	18	6	24	

The p-value (0.80) indicates no significant difference in Co-Amoxiclav resistance between GABHS and other pathogens.

During the 15-month study period, tonsillitis cases peaked in March and April. Two cases involving *Candida* spp. were identified, both in children under 10 years of age; these were excluded from the study. Additionally, out of 280 cases, 120 patients (42.86%) had used antibiotics before visiting the hospital, with azithromycin being the most commonly used 47(16.78%). Patients who had taken Co-Amoxiclav and Amoxicillin were excluded from the study but other commonly used antibiotic received patient were included like Azithromycin, Cefixime, Cefuroxime, Cefpodoxime and etc. These findings suggest a potentially higher rate of over-the-counter antibiotic use than captured in this study

## DISCUSSION

Acute tonsillitis is a common and debilitating condition worldwide. This study focused on its clinical presentation, throat swab results, and trends in antibiotic sensitivity in western Nepal. The findings highlight the primary bacterial pathogens responsible for the condition, their antibiotic sensitivity patterns, and current resistance trends, which are crucial for ensuring effective treatment.

The mean age of patients in this study was 26.51 years, with the highest prevalence of tonsillitis in the 21–30 years age group. This finding is

consistent with previous studies regarding mean age [3,6]. However, the most prevalent age group in other studies was children under 10 years [2,3], which differs from our findings. This discrepancy may be due to the exclusion of children under 5 years in our study, as throat swab collection in this age group was anticipated to be challenging. The gender distribution in our study (53.2% female, 46.8% male) aligns with some studies [3], suggesting that acute tonsillitis affects both genders equally [4,7]. However, other studies report a higher prevalence in males [8]. These variations may reflect differences in study populations, environmental factors, or healthcare-seeking behaviors across regions.

Throat discomfort, including dysphagia, odynophagia, and irritation (93%), emerged as the most common symptom, followed by fever, constitutional symptoms, and neck swelling. These findings align with other studies [9,10]. Acute superficial tonsillitis (50.7%) was the most common type in our study, differing from other studies where acute parenchymatous (61%) [9] or acute follicular tonsillitis (65%) [10] were more prevalent. These variations may be due to differences in sociodemographic regions. Enlarged and tender lymph nodes were observed in 62.5% of patients, consistent with some studies [11], while others report higher rates (up to 80.88%) [12]. Increased lymph node involvement may be associated with seasonal flu and fever. Throat swabs were positive in only 60 patients (21.4%), with the remaining cases

showing no growth or normal flora. This result is significantly lower compared to similar studies [3], likely due to the overuse of antibiotics in our region. GABHS was isolated in 36 samples (12.9%), which is also lower than other studies reporting isolation rates of 25–50% [1–3]. Similar findings were reported in Nigeria (15–30%) [13] and Singapore (23%) [7]. These variations may be attributed to differences in treatment protocols, geographical factors, timing of sample collection only acute phase shows high bacterial load, laboratory factor such as proper handling of specimen, incubation and processing and Patients on carrier state. In our study, acute membranous tonsillitis showed the highest bacterial growth (10.35%), lower than another study reporting 31.2% GABHS isolation in membranous tonsillitis [14].

Among the 60-throat swab-positive samples, 36 were GABHS. The sensitivity of GABHS to Co-Amoxiclav was 77.77%, compared to 85.7% in Heussien et al. (2020) [15] and 59.6% in Abraham et al. (2019) [3]. Nabipour et al. (2005) reported 87.7% resistance to plain amoxicillin [4]. These varying patterns of antibiotic sensitivity and resistance highlight the influence of time, location, and antibiotic treatment protocols.

A significant proportion of patients (42.9%) had already received antibiotic treatment before presenting to the hospital, with azithromycin being the most commonly used (16.4%). Patients who had taken Co-Amoxiclav were excluded

from the study. This suggests a potentially higher rate of over-the-counter antibiotic use than captured in our study. Similar findings were reported by Ughasoro et al. (2021), with 35.6% of patients having prior antibiotic use [13]. This practice is common in underdeveloped and developing countries, likely due to lax antibiotic prescribing policies and easy access to medications over the counter.

Our study has a few limitations. The lack of anaerobic bacterial cultures may have resulted in the omission of some pathogens, and many untested antibiotics limited the evaluation of alternative treatment options. Convenience sampling methods may have introduced selection bias. As a single-center study conducted in a tertiary care hospital, the external validity of our findings cannot be generalized. Larger, multicenter cohort studies with a longitudinal design are needed to validate these results and further investigate potential causal relationships.

## CONCLUSIONS

The current study found a low throat swab culture positivity rate (21.4%), with GABHS being the most prevalent bacterial pathogen (12.9%). Co-Amoxiclav demonstrated 77.77% sensitivity to GABHS. However, the declining sensitivity pattern in recent years underscores the need for judicious antibiotic use and the exploration of alternative antimicrobial therapies.

## CONFLICT OF INTEREST

None

## SOURCES OF FUNDING

None

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