

BACTERIOLOGICAL EVALUATION OF SURFACE AND CORE TONSILLAR TISSUE IN CHRONIC TONSILLITIS

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ABSTRACT

Background: Chronic tonsillitis results from recurrent acute tonsillitis which is thought to result from insufficient penetration of antibiotics into the core of the tonsil or inappropriate antibiotic therapy. In tonsillitis, bacterial evaluation of both the tonsillar surface and core reveals that surface swabs may not always accurately reflect the pathogens present in the deeper tonsillar tissue and comparing the surface and core microflora can help the otorhinolaryngologist to determine the etiological agent and initiate effective treatment and also reduce the emerging and impending antibiotic resistance. **Materials and Methods:** Twenty patients of both genders with diagnosis of chronic tonsillitis and with ASO titres positive were included in the study. The swabs obtained from surface of the tonsil before surgery and from the core of tonsil after tonsillectomy were cultured and antibiotic sensitivity testing was done for pathogens. **Result:** Group A beta-hemolytic streptococci (GABHS) (32.5 %) was the most common pathogen isolated from both surface and core followed by *Staphylococcus aureus* (20%), *Streptococcus pneumoniae* (7.5 %) and rest of the bacteria isolated were *Klebsiella pneumoniae*, *Pseudomonas* (7.5%). The antibiotic sensitivity testing yielded sensitivity to amoxycillin and clavulanic acid combination, ciprofloxacin, cotrimoxazole and Levofloxacin. Surface swab culture does not always represent the bacteriology of the core. **Conclusion:** The role of throat swabs in the management of chronic tonsillitis is doubtful. Tonsillar deep tissue cultures may help clarify tonsillar microbiology and guide the treatment of patients with chronic tonsillitis.

INTRODUCTION

Adenoid and tonsils which are parts of Waldeyer's ring play a protective role against infections in healthy individuals and they may become a reservoir for pathogenic microorganisms. Chronic tonsillitis results from recurrent acute tonsillitis which is thought to result from insufficient penetration of antibiotics into the core of the tonsil or inappropriate antibiotic therapy.^[1]

Tonsil infection may occur primarily or secondarily as a result of upper respiratory tract infections commonly preceded by viral infections and they include Adenovirus, Epstein Barr Virus (EBV), Influenza virus and Echovirus and the common bacteria involved is Group A haemolytic streptococcus (GABHS).^[2] Effective treatment of patients with tonsillitis depends on knowledge of the

infecting organisms. In tonsillitis, bacterial evaluation of both the tonsillar surface and core reveals that surface swabs may not always accurately reflect the pathogens present in the deeper tonsillar tissue.^[3] Specifically, core tissue samples often harbour different or more diverse bacterial species than surface swabs, highlighting the limitations of relying solely on surface cultures for diagnosis and treatment of tonsillitis. However, for centuries there has been discussion on the presence and identity of micro-organisms on the surface and in the core of the tonsils, and their role in causing tonsillitis, as studied in different settings without evidence for specific antibiotic treatment.^[4,5] Thus, comparing the surface and core microflora can help the otorhinolaryngologist to determine the etiological agent and initiate effective treatment and also reduce the emerging and impending antibiotic resistance.

This study was aimed to determine the correlation of bacteria on the surface and core tonsillar tissue and also to predict bacterial sensitivity to some antibiotics.

MATERIALS AND METHODS

The present study was a prospective analytical study conducted in the Department of Otorhinolaryngology at a tertiary care center in South India after obtaining Institutional Ethics Committee approval (Lr.No.47/2020 dated 07/09/2020). The study included twenty patients of both genders aged between 12 and 40 years and attending the Department of Otorhinolaryngology OPD. Patients with different grades of tonsillar enlargement meeting the criteria for diagnosis of chronic tonsillitis with serum positive for ASO titers were included in the present study. Data was collected from all the subjects in a standardized proforma which included details of age, gender, clinical history, complete ENT and head and neck examination, blood investigations like CBC, Blood grouping and Rh typing, Bleeding time, clotting time, prothrombin time, ASO titer, Chest x-ray, renal functional tests. All the patients were admitted one day before for surgery with suitable antibiotic coverage for a period of 7 days. Collection of Swab for microbiological examination and antibiotic sensitivity testing:

1. On the day of surgery before intubation, three swabs were taken trans-orally under direct vision from the surface of the tonsils without touching other parts of the oral cavity and oropharynx, and was placed in a transport medium.
2. Following this, tonsillectomy was performed by dissection and snare technique. Immediately after excision, the tonsil was dipped in povidone iodine solution for 30 – 45 seconds. Then it was rinsed in sterile saline solution and sectioned into two pieces under strict aseptic conditions. The same procedure of rubbing a sterile swab was applied to the core of the excised tonsils, avoiding its outer surface, and placed into a transport medium.

Processing of specimen: In the microbiology laboratory, the surface swabs and swabs from the core tissue were processed according to the standard microbiological techniques.

1. One swab was used for the smear preparation and the smear stained by Gram's method and observed under microscope for the presence of bacteria and its morphology, Gram's reaction, and findings were noted.
2. The second swab was used to inoculate on blood agar, chocolate agar, McConkey agar. The plates were incubated at 37°C, blood agar and chocolate agar incubated at 37°C in a 5-10% CO₂ atmosphere for a period of 24 hours and afterward colonies were observed for bacteria grown on that is identified by using standard microbiological conventional techniques.

3. The third swab was kept in BHI broth and incubated at 37°C as a measure of backup culture if there is no growth on solid media with the second swab. Subcultures were done from this broth culture onto blood agar, chocolate agar McConkey agar and incubated at 37°C for 24 hours and observed for the presence of bacterial growth. If there is growth, they are identified by standard microbiological conventional techniques.
4. Antibiotic sensitivity test was performed by the Kirby-Bauer Disk Diffusion method by measuring the zone of inhibition around the antibiotic discs and reference tables were used to determine if the bacteria are sensitive (S), intermediate (I) or resistant (R) to the antimicrobial drugs.

Statistical analysis: Continuous variables were expressed as mean \pm standard deviation (SD). Frequency was expressed as number (percentage), (n (%)). The difference in the means of type of bacteria was analysed by independent samples t-test. Data was analysed using Microsoft excel spread sheets and Statistical Package for Social Sciences (SPSS, Inc., Chicago IL) for windows version 16.0. A p-value less than 0.05 was considered as statistically significant.

RESULTS

A total of 20 patients of both genders were recruited for the study and the mean age of the patient was 23.65 \pm 5.2 years. The patients fall in the age range of 11-20 years in 5 (25%), between 21-30 years in 13(65%) followed by 2 (10%) patients in 31-40 years range and majority of the patients (n=13) belonged to 21-30 years range. 60% (n=12) of the patients are of female gender selected for study. Out of 20 patients, who were included in the present study there was grade III enlargement of tonsils in 13(65%) patients followed by grade II in 25 % of the patients.

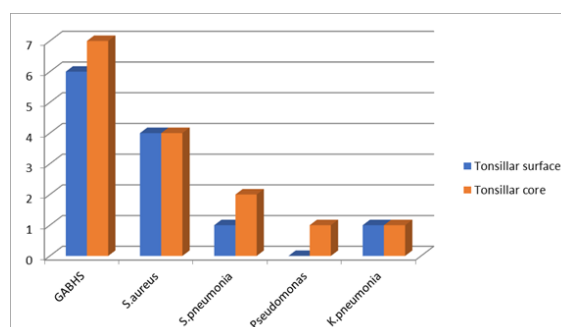


Figure 1: Comparison of organisms isolated from tonsillar surface and core of tonsillectomy specimen

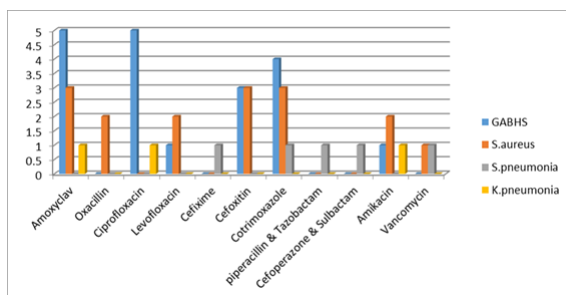


Figure 2: showing the antibiotic sensitivity from the surface swab

Table 1: Discrepancy between organism isolated from surface and core

Cases	Surface organism	Core organism	Surface core discrepancy
Case 1	S.aureus	GABHS	Yes
Case 2	S.aureus	GABHS	Yes
Case 3	Normal flora	S.pneumonia	Yes
Case 4	S.aureus	S.aureus	No
Case 5	Normal flora	S.aureus	Yes
Case 6	Normal flora	GABHS	Yes
Case 7	S.pneumonia	K.pneumonia	Yes
Case 8	K.pneumonia	Normal flora	Yes
Case 9	K.pneumonia	Normal flora	Yes
Case 10	Normal flora	S.pneumonia	Yes
Case 11	No culture	No culture	No
Case 12	GABHS	S.aureus	Yes
Case 13	S.aureus	GABHS	Yes
Case 14	GABHS	Normal flora	Yes
Case 15	GABHS	Normal flora	Yes
Case 16	GABHS	S.aureus	Yes
Case 17	Normal flora	GABHS	Yes
Case 18	GABHS	GABHS	No
Case 19	GABHS	Pseudomonas	Yes
Case 20	GABHS	GABHS	No

In this study, five different pathogens were isolated. GABHS (group A beta-hemolytic streptococci) was the most common isolate found in 13(32.5%) cases, followed by Staphylococcus aureus in 8(20%), Streptococcus pneumonia in 3(7.5%), Klebsiella pneumonia in 2(5%), and Pseudomonas in one case (2.5%). [Figure 1] is showing the organisms isolated from surface of the tonsils and core of the tonsillectomy specimens.

[Table 1] is showing the discrepancy between the organism isolated from surface of the tonsils and from core of tonsillectomy specimens. The discrepancy was found in 80%(n=16) of the microbiological reports. In the present study, 2(10%) cases showed the same pathogen from the tonsillar surface as well as from the core and in 7(35%) cases, different pathogens were isolated from the surface and core. In one case, normal flora was isolated from both surface of the tonsil and core of tonsillectomy specimen. The antibiotic susceptibility was showing [Figure 2] the susceptibility of bacteria for amoxycillin and clavulanic acid combination, ciprofloxacin and cotrimoxazole.

DISCUSSION

Chronic tonsillitis is a very common disease affecting the palatine tonsils in children and empirical treatment of this condition is with the antibiotics

which target the common pathogen GABHS and in some cases the antibiotic may be inappropriate or insufficient where surgery becomes inevitable. Tonsillectomy is the most common surgical procedure performed in cases of chronic tonsillitis or in recurrent tonsillitis. To avoid surgery, surface swab cultures and antibiotic sensitivity may help but still these measures may not help which results in chronicity and subsequent surgical intervention is needed. To know these discrepancies and to avoid surgical intervention the core of tonsillectomy specimen cultures may help in evidence-based treatment for chronic tonsillitis.^[6,7] If the surface swab pathogen is representing the core pathogen, then antimicrobial therapy based on the surface swab would be reliable.^[8] There is evidence that the pathogenic organisms were found in both surface and core tissues of patients with tonsillitis.^[9] Hence, this study was conducted to compare the microflora of tonsillar surface and core tissue in those who are undergoing tonsillectomy.

In our study, majority of patients belonged to 21-30 years and 60 % were of female gender with chronic tonsillitis as the major indication for tonsillectomy. Similar observations with female predominance were noted in studies conducted by Kurien et al,^[10] and Sriram et al.^[11] In the present study, grade 3 tonsillar hypertrophy was found in 65%, grade 2 in 25%, and grade 4 in 10% of cases. The same observations were

reported in a study conducted by Manandhar et al,^[12] as grade 3 hypertrophy in 72%, grade 2 in 24%, grade 4 in 14%. Abhaykumar et al,^[8] noticed that chronic tonsillitis was the common indication for tonsillectomy in 84% and tonsillar hypertrophy in 16% of isolates.

Five different pathogenic organisms have been isolated from 20 patients in our study, in which GABHS was the most common organism (35%), followed by *Staphylococcus aureus* (20%), *Streptococcus pneumoniae* (7.5%), *Klebsiella pneumoniae* (5%), *Pseudomonas* (2.5%). The current study showed 32.5% of tonsillar surface culture with a pathogenic organism compared to 37.5% in tonsil cores. Similar findings were noted in Kurien et al,^[10] study with 55% on the surface and 72.5% in the core and Shishegar et al,^[9] study showed 76% on the surface and 79.6% of organisms in the core. GABHS was isolated in our study which was consistent with other studies conducted in Indian population. There were discrepancy in surface and core cultures in our study which was also noticed in other studies.^[9,10] In the present study, GABHS was the commonest organism cultured in both tonsillar surface and core. In surface culture, GABHS showed sensitivity to Amoxi-clav in 5 isolates, Ciprofloxacin in 5, Cefoxitin in 3, Cotrimoxazole in 4 isolates and in core culture, GABHS showed sensitivity towards Ciprofloxacin in 5 isolates. Amoxi-clav, Cotrimoxazole 4 isolates each. Levofloxacin in one isolate.

From this study, it was evident that the routine surface swab samples in patients with chronic or recurrent tonsillitis were neither reliable nor valid. If the core organisms were identified, the antimicrobials can be selected appropriately and the need for elective tonsillectomy could be obviated. Some studies, show that core organisms could be identified preoperatively by core FNA sampling and found to be reliable with dissected core sampling.^[9]

Limitations of the study: In the present study sample size was small which may not represent the significant population. Prior antibiotic usage was not considered during this study. To determine the pathogen or part of normal flora causing the disease, it would be useful to have a control group like those who are undergoing tonsillectomy for a noninfective cause. Hence, further studies should be done to overcome these issues.

CONCLUSION

The role of throat swabs in the management of chronic tonsillitis is doubtful. In view of our study and other studies investigating tonsillar bacteriology, it is obvious that surface culture does not reliably predict core pathogens in cases of chronic infection. *Staphylococcus aureus* and GABHS were the most common isolated organisms from core and surface of the tonsil followed by *S. pneumoniae*, *Klebsiella pneumoniae* and *Pseudomonas* and for effective treatment, the antibiotics sensitive against these pathogens like Amoxycillin-Clavulanic acid, Ciprofloxacin, Cefpodoxime, Amikacin, Levofloxacin should be chosen. Thus, tonsillar deep tissue cultures may help clarify tonsillar microbiology and guide the treatment of patients with chronic tonsillitis.

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