



## Comparative Study of Surface and Core Microflora of Chronic Tonsillitis

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### Abstract

*The main objective of this study is to find the correlation and differences between the microflora isolated and characterized from the surface and core regions of the tonsils, where the subjects were initially defined as tonsillitis. The maximum age incidence was from 6 to 12 years where the sex incidence was more in females than males. In majority of the patients, jugulodigastric nodes are palpable where the squeeze test was positive to 22% of cases. Sore throat and odynophagia are the symptoms predominantly found and these were presented with 2 to 3 years of duration among maximum cases, where in the average duration was 1 year and 7 months. Majority of the patients had chronic parenchymatous tonsillitis but enlarged adenoids were the most commonly associated ENT condition. Among the bacterial isolates, 87% was possible to aerobic isolation whereas 13% possibility was for both aerobic and anaerobic bacterial members. In the surface region,  $\beta$  haemolytic streptococci and *Staphylococcus aureus* are predominant isolates whereas in core region,  $\beta$  haemolytic streptococci and *Streptococcus pneumoniae* are possible. The variations in the bacterial isolates were possible maximum in core region compared to surface. *Bacteriodes* was the common anerobe found in both core and surface but dominance of *Fusobacterium* was identified in surface area only. The raised ASO titre was found in majority of the patients. Norfloxacin and ciprofloxacin are the common and routine antibiotic classically found effective against the isolates that re-establishing the superiority of the newer generation antibiotics to traditional penicillin.*

**Keywords:** Chronic tonsillitis, Bacteriological investigation, surface, core, comparativeness.

### Introduction

Chronic tonsillitis is the most common disease in throat that occurs predominantly in the younger age group<sup>1</sup>. The disease is diagnosed mainly by the history and clinical examinations. Superficial

tonsillar swabs are often used as a guide in identifying the offending pathogenic organism and the proper selection of therapy in acute and recurrent tonsillitis<sup>2</sup>.

In recurrent tonsillitis, the tonsil core harbours numerous bacteria, some of which are pathogenic and may occur in great numbers. The most frequent bacteria are *Haemophilus influenzae*, followed by *Staphylococcus aureus* and *Streptococcus pyogenes*. A high tissue concentration of these bacteria correlates with clinical parameters of infection and hyperplasia of the tonsils. The role of *Streptococcus pyogenes*, while undisputed in acute and epidemic tonsillitis, should perhaps be re-evaluated in recurrent and/or hyperplastic tonsillitis<sup>3</sup>. Adenotonsillar disease can be divided into two categories, infection and hypertrophy. They are closely related with low grade infection causing hypertrophy in most instances<sup>1,2</sup>.

Minor advances have been accomplished in the treatment of infectious processes, mainly the advent of new antibiotics. More important advances have occurred in the field of airway obstruction secondary to adenotonsillar hypertrophy. Currently there exists a significant body of knowledge on obstructive sleep apnea. With improvement in anesthetic and surgical techniques younger children are being treated earlier to relieve airway obstruction<sup>4</sup>.

Antimicrobial treatment is often not successful to completely cure the pathogens and prevent recurrences of the infections commonly found in the tonsils<sup>3,4</sup>. In most of the cases, failure to control or completely remove the microbial load in the core of tonsils either due to inappropriate antimicrobial therapy or inadequate antibiotic penetration in the core paves way to either persistence of core infections or reinoculation of the initially sterilized surface. In some situations, the surface culture is be the source for the infections in core, if that is the case, then rational treatment could be directed to the organisms cultured by surface swab<sup>5</sup>.

Tonsillectomy is indicated in recurrent acute tonsillitis for at least two years with five or more acute attacks per year, the procedure may be associated with transient bacteraemia in some cases which may or may not be associated with

any enhanced post-operative morbidity<sup>6,7</sup>. The incidence of bacteraemia associated with tonsillectomy varies among different investigators<sup>6,7,8</sup>.

Although many bacteriology studies on tonsillar diseases have been completed, all have been confined to children and were characterized by a paucity of cases<sup>9</sup>. The purpose of this study was to analyze the underlying bacterial pathogens in tonsillar disease of both surface and core region. Some studies have concluded that the determination of the surface flora alone is not useful in predicting the infectious state. Thus comparing the surface and core microflora provide clear ideas in determining the etiological agent and to initiate treatment successfully.

### Materials and Methods

This is a retrospective study undertaken in 100 consecutively operated tonsillectomy cases at Rajah Muthiah Medical College, Annamalai University, Annamalai Nagar. The patients under ENT complete otorhinolaryngological examination and their ages ranged from 4 to 30 years. About 34 cases were males and 66 were females. The study was approved by institutional ethical committee and all the subjects included in this study were given their written consent as a part of ethical consideration.

The duration of symptoms ranged from 2 to 3 years of duration among maximum cases, average duration was 1 year and 7 months. Most of them had received antibiotic prior to our evaluation, but none of the case in the study had any antimicrobial therapy for at least 1 week prior to surgery. After the patient was intubated a tonsillar surface swab was obtained by rotating a sterile cotton wool swab over the surface of the tonsil not touching other part of oropharynx<sup>10,11,12</sup>. Following this tonsillectomy was performed by dissection technique.

Immediately after excision, the tonsil was dipped in povidineiodine solution for 30 seconds and then it was rinsed in sterile saline solution further sectioned into two pieces under strict aseptic

condition. The same procedure of rubbing a sterile swab was applied to the surface of excised tonsils avoiding its outer surface. These were then transferred to the bacteriology laboratory. The specimen was cultured on 5% sheep blood agar, chocolate agar and in brain heart infusion agar for anaerobic cultivation<sup>11,12</sup>. The plates were incubated at 37°C in the presence of 5-10% CO<sub>2</sub> for 24 to 48 hours. Subculture from Robertson's cooked meat medium has been also done. Colony identification was accomplished using the standard technique. Biochemical confirmations were also done according to the standard bacteriological methods.

Growths of  $\beta$  haemolytic streptococci, *Streptococcus pneumoniae*, *Staphylococcus aureus*, *Enterococcus spp*, *Klebsiella spp*, *Proteus spp*, *Fusobacterium*, *Peptostreptococcus* and *Bacteriodes* on any of the media were identified by standard methods.  $\beta$  hemolytic streptococcal (BHS) colonies could not be classified further into their Lancefield groups because of non availability of this facility<sup>12</sup>. The sensitivity of the isolates was tested against seven common antibiotics, using Himedia antibiotic discs.

## Results

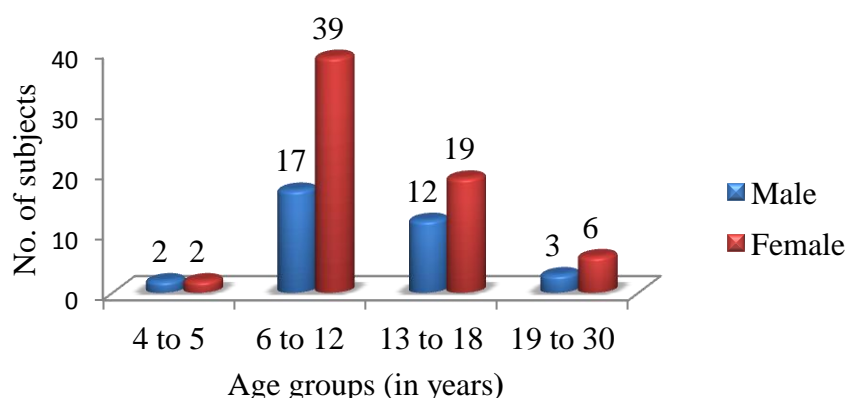
The commonest age of occurrence is between 6 and 12 years accounting 56 cases (56%). The minimum age of the patient was found between 4 to 5 years (4 cases). Female predominance was

found among the cases where 34 cases were males and 66 were females. The age wise and sex distribution of cases subjected to this study was impregnated in figure 1. Majority of the patients belonged to low socio economic status (87%). In clinical examinations, the palpability of jugulodiaphragmatic nodes are found among 72 cases only. Further the squeeze test was positive among 22 cases and others showed negative.

The indication for tonsillectomy was ascertained by detailed history and physical examinations (Table 1). Recurrent tonsillitis, i.e., two or more episodes of tonsillitis for two or more consecutive years was an indication for tonsillectomy in 94 cases. The second most important indication for tonsillectomy which appeared in the study was tonsillar size 3+/4+ denoting 50-70% and greater than 75% obstruction of the airway, respectively. The chief complaint relating to the hypertrophy of tonsil was mainly mechanical, either leading to obstruction in airway or deglutition problem.

This was an indication for tonsillectomy (inclusive of other related factors) in 26 cases. Obstructive sleep apnoea syndrome (OSAS) characterized by episodes of apnoea during sleep was indicated in two cases. This was considered an absolute indication for tonsillectomy. General ill health and failure to thrive was in itself not considered important for surgery, but was a co-existing factor in number of cases.

**Figure 1:** Age and sexwise distribution of subjects (n=100)



**Table 1:** Indications of tonsillectomy versus age groups

Age groups (in years)	Indications of tonsillectomy		
	Recurrent tonsillitis (n=94)	Tonsillar size (n=26)	General ill health (n=16)
4 -5	3 (3.2)	1 (3.8)	1 (6.2)
6 – 12	54 (57.4)	15 (57.7)	2 (12.5)
13 – 18	30 (32)	8 (30.8)	4 (25)
19 – 30	7 (7.4)	2 (7.7)	9 (56.3)

[Figure in parenthesis denoted percentage of indications of tonsillectomy]

The tonsillar surface culture is compared with interior tonsillar core and the data was depicted in table 2. Percentage of patients had differential correlation between organisms isolated from two

sites. The samples in both surface and core showed aerobic isolation whereas anaerobic isolation was possible only to 16 and 13 subjects respectively (Table 2).

**Table 2:** Distribution of aerobic and anaerobic bacterial isolates

Type of isolates	Number of samples	
	Surface samples (n=100)	Core samples (n=100)
Aerobic bacteria	89	94
Anaerobic bacteria	16	13

Overall surface culture was in variance as to the presence or absence of core pathogens and it was identical. Surface pathogens differing from core pathogens occurred in all cases. The bacteria isolated according to prevalence in decreasing order of frequency were Group beta haemolytic

streptococci followed by *Staphylococcus aureus*, *Streptococcus pneumoniae*, *Klebsiella pneumoniae*, *Proteus sp*, *Enterococcus sp*. The order of isolation was vary from surface to core tonsillitis specimens (Table 3).

**Table 3:** Comparison of bacterial pathogens isolated from surface and core tonsils

Types of the bacterial isolates	Number of samples supported	
	Surface specimen of tonsils (n=100)	Core specimen of tonsils (n=100)
Aerobic bacterial isolates		
β haemolytic streptococci	40	32
<i>Staphylococcus aureus</i>	21	9
<i>Klebsiella sp</i>	19	21
<i>Streptococcus pneumoniae</i>	12	24
<i>Proteus sp</i>	5	10
<i>Enterococcus sp</i>	2	4
Anaerobic bacterial isolates		
<i>Bacteriodes sp</i>	7	5
<i>Fusobacterium sp</i>	6	4
<i>Peptostreptococcus sp</i>	3	4

Next to β haemolytic streptococci, *Staphylococcus aureus* and *Streptococcus pneumoniae* are dominated in surface and core specimens respectively. Five and seven cases of aerobic and anaerobic polymicrobial infections were

encountered in the study. There is no statistical significant correlation between the type of bacteria isolated and frequency of tonsillitis in the preceding year and/or any other clinical manifestations (Table 3).

**Table 3:** Test of significance and Z value for equality and proportions among isolates

Types of the bacterial isolates	Statistical correlation	
	Significance	Z value
Aerobic bacterial isolates		
$\beta$ haemolytic streptococci	Non Significant	1.24
<i>Staphylococcus aureus</i>	Significant	2.28
<i>Klebsiella</i> sp	Non Significant	0.44
<i>Streptococcus pneumoniae</i>	Significant	2.59
<i>Proteus</i> sp	Non Significant	1.36
<i>Enterococcus</i> sp	Non Significant	0.85
Anaerobic bacterial isolates		
<i>Bacteriodes</i> sp	Non Significant	0.54
<i>Fusobacterium</i> sp	Non Significant	0.59
<i>Peptostreptococcus</i> sp	Non Significant	0.50

In this study, ASO titre was raised among 75% of the cases. The correlation of  $\beta$  haemolytic streptococci to ASO titre showed 99.9% sensitive (among 50  $\beta$ HS, 57 showed serologically positive to ASO). With the view to examine the bacterial etiology of tonsillitis, the antibiotic sensitivity patterns showed sensitive to antibiotics and two way analyzing of variance has been carried out

and the detailed description of the sensitivity pattern was depicted in table 2. Further there is a no significant difference in the sensitivity of different types of bacteria exposed to each of the antibiotics in the surface of tonsils. So, the overall, the antibiotic sensitivity pattern is not showing more significance in difference between the surface and core of tonsils (Table 4).

**Table 4:** Test of significance and Z value for equality and proportions among sensitivity pattern

Antibiotics	Statistical correlation	
	Significance	Z value
Amikacin	Significant	1.99
Ciprofloxacin	Non Significant	1.31
Cotrimoxazole	Non Significant	0.31
Gentamycin	Non Significant	0.59
Norfloxacin	Significant	2.53
Penicillin	Significant	42.4
Tetracycline	Non Significant	0.42

## Discussion

This study showed that *Staphylococcus aureus* and *Streptococcus pneumoniae* are the most common pathogenic bacteria cultured both in adults and children next to  $\beta$  haemolytic streptococci. Most of the studies highlighted that *Pseudomonas aeruginosa* is a common causative organism in recurrent tonsillitis<sup>10</sup> but in this study even single sample does not support that. Our knowledge of bacteria of the tonsils has very largely been obtained from the study of swab cultures and smears made directly from the

surface of tonsils *in situ*. This surface is necessarily contaminated by bacteria from the mouth, throat, nose, food, inspired air and such examinations may not only give very correct data about the bacteria that actually exist in the depths of the tonsils but also may at least partly explain the reason why such a complex bacterial flora has been attributed to this organ<sup>13</sup>.

The study indicates that the routine surface culture obtained from the tonsil do not always represent the nature of the bacterial flora of the core of the tonsil. Recurrent attacks of tonsillitis make the wisdom of using frequent course of broad



spectrum antibiotics questionable. Surgical excision of tonsil is the best modality of treatment for recurrent infection, as pathogens from core of the tonsil remain unidentified and are resistant to the antibiotic therapy based on the tonsillar surface microflora<sup>2,3,12</sup>.

## Conclusion

Since the mechanism of activation of infection in recurrent tonsillitis is unknown, knowing the bacteriology does not help us to treat the disease. However, it may be a stepping stone to eventually understanding whether the bacteria play a role in reactivating recurrent infections. From previous and current studies, there is no relationship between bacteriology and recurrent infections. Thus exact identification of bacterial isolates in tonsillitis and their antibiotic sensitivity pattern could revolutionize the management of chronic tonsillitis.

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