Association between adenoid bacteriology and clinical characteristics of adenoid-related diseases in children

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Abstract
Introduction: The aim of this research is to find the association between the pathogenic bacteria obtained from the adenoid culture and clinical characteristics of adenoid-related diseases in children.

Methods: In this retrospective study, we reviewed the medical records of children who had adenoidectomy for adenoid-related diseases. Demographic data, diagnoses, indications for adenoidectomy and bacterial culture results were collected. The adenoid size was measured in the lateral skull X-ray as adenoid–nasopharyngeal ratio. Associations between the culture results and the demographic data, adenoid size, and the diagnoses were analyzed.

Results: There were 407 children who had adenoidectomy for obstructive sleep-disordered breathing (75.2%), otitis media with effusion (19.2%), and chronic sinusitis (5.6%). Median age was 5.9 years. Common pathogenic bacteria in the adenoid were Haemophilus influenzae (26.2%), Staphylococcus aureus (23.5%), Streptococcus pneumoniae (18.2%), and Moraxella catarrhalis (12%). The patient’s age had significant association with the prevalence of pathogenic bacteria. S. pneumoniae was most prevalent in young children up to 7 years. S. aureus was more common in children over 7 years. H. influenzae had similar prevalence in all age groups. Size of the adenoid and type of adenoid-related diseases had no association with the outcome of bacterial culture.

Conclusion: Age of the patients was the significant factor associated with the bacteriological findings of the adenoid while size and types of adenoid-related diseases were not associated with the outcome of bacterial culture.

Keywords
Adenoid, adenoidectomy, bacteriology

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Introduction
The adenoid is a part of the Waldeyer’s ring of lymphoid tissue in the nasopharynx which is the gateway to the upper respiratory tract in children. The adenoid has a protective function against the organisms entering the upper respiratory tract via the nasal cavity. The lymphoid tissue in the adenoid is stimulated by the exposure to antigen or bacteria, leading to lymphoid proliferation, production of immunoglobulins, and cytotoxic activity.¹ Inflammatory reaction and bacterial infection of the adenoid are the causes of adenoid hypertrophy and the adenoid become the reservoir of bacteria in the upper respiratory tract. The Eustachian tube connects the nasopharyngeal cavity to the middle ear. Therefore, the adenoid involves in the pathology of the diseases of the nose, sinus, and the middle ear. Adenoid hypertrophy is the cause of obstructive sleep-disordered breathing

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(OSDB), and adenotonsillectomy is the definite surgical management for pediatric obstructive sleep apnea (OSA). Bacteria in the adenoid are the underlying causes of pediatric sinusitis and otitis media with effusion (OME). Bacteriology of the adenoid was found with similarity to the pathogenic bacteria in the middle meatus and middle ear effusion. Adenoidectomy is a part of the surgical management of pediatric sinusitis and otitis media according to the clinical practice guideline. Factors for consideration of adenoidectomy include the age of the children, diagnosis of adenoid-related diseases, and the size of the adenoid. There was a suggestion for future study to investigate the differences in adenotonsillar microbiome according to the demographic data and the clinical presentation of adenoidal diseases. Association of the specific bacteria in the adenoid and type of adenoid-related diseases have not been established. The aim of this study is to review the bacteriology of the adenoid from the patients who had adenoid-related diseases and to find the association between the pathogenic bacteria and the clinical characteristics of the patients including age, adenoid size, and the diagnosis.

Methods

This research was a retrospective, descriptive study. We collected the data from the medical records of pediatric patients admitted for adenoidectomy or adenotonsillectomy between August 2005 and December 2018. The justification of the sample size was based on the proportion of adenoid-related diseases from our previous study in 2004. A power analysis was based on the prevalence of Haemophilus influenzae, which was the most common bacteria in adenoidal diseases from the study of Brodsky and Koch and the study of Shin et al. The sample size calculated was 401 cases for a two-sided study with 0.01 significance level and a power of 90%. Inclusion criteria were the patients who had adenoidectomy or adenotonsillectomy for adenoid-related diseases. Exclusion criteria were the patient with incomplete data in the medical records. The institutional review board of the Faculty of Medicine Siriraj Hospital approved the research.

Data gathering included demographic data, clinical presentation, diagnosis of adenoid-related diseases, indication for surgery, and results of the adenoid culture. The adenoid size was evaluated by lateral skull film and measured in an adenoid–nasopharyngeal ratio (ANR) by the Fujioka method. The surgery and adenoid culture were done in the same pattern in all cases. Adenoidectomy was done using an adenoid curette, and fresh specimen of the adenoid was sent to the microbiological laboratory for bacterial culture within 30 min. The specimen was cut into small pieces and inoculated onto chocolate agar, 5% sheep blood agar, and MacConkey agar plates for the cultivation of aerobic and facultative anaerobic bacteria. Gram staining from adenoid tissue was also carried out. The inoculated plates were incubated at 35°C aerobically (MacConkey) and under 5% carbon dioxide (chocolate agar and 5% sheep blood agar plates), and examined after 24, 48, and 72 h of incubation. The growth of suspected bacterial pathogen was identified by Gram staining, conventional biochemical method, and automated VITEK 2 system. Antimicrobial susceptibility testing by disk diffusion method was performed according to CLSI guidelines. The culture results were recorded.

Statistical analyses

Descriptive statistics were used for demographic and descriptive data. Types of specific pathogenic bacteria from adenoid culture were analyzed in association with the patient’s age, the size of the adenoid, and the diagnosis of adenoid-related diseases. Chi-square and Fisher’s exact test were used for univariate analysis. Logistic regression was used for multivariate analysis between clinical features and types of bacteria from the adenoid culture. Statistical analysis was done using SPSS version 22.

Results

The data of 407 pediatric patients were included in the study. Demographic data and clinical characteristics of the patients were described in Table 1. The median age was 5.9 years with interquartile range (IQR) 3.7. The most common diagnosis of adenoid-related diseases was OSDB, followed by OME and chronic sinusitis. There were 86 (21.1%) patients with more than one diagnosis and 8 patients (2%) had all three diagnoses.

Investigation of the patients before adenoidectomy was lateral skull X-ray, polysomnography in cases with OSDB, and hearing test in patients with OME. The results were shown in Table 1. ANR was more than 0.7 in 78.2% of the patients.

Adenoid bacterial culture was shown in Table 2. The culture was positive in 400 cases and no growth in 7 cases, with 755 bacterial isolates. H. influenzae was the most common organism found in the adenoid culture and beta-lactamase-negative isolates were found more than beta-lactamase-positive ones (20.9% and 4.8%, respectively). Staphylococcus aureus was the second most common bacterial pathogen; only one case of methicillin-resistant strain was found in this series. Streptococcus pneumoniae was the third most common organism; penicillin-susceptible isolates were found more commonly than penicillin-resistant ones (17.9% and 0.3%, respectively). Beta-lactamase-negative Moraxella catarrhalis was found more than beta-lactamase-positive isolates (11.4% and 0.5%, respectively).

Adenoid bacterial culture of each diagnosis was shown in Figure 1(a)–(c). The pattern of prevalence was similar in OSDB, OME, and chronic sinusitis. Figure 2 showed the percentage of bacteria in patients with single diagnosis and multiple diagnoses of adenoid-related diseases. There was more prevalence of Pseudomonas aeruginosa in patients.
with multiple diagnoses with statistical significance ($p = 0.04$). There were two cases of patients with multiple diagnoses who had penicillin-resistant *S. pneumoniae*, while none was found in the patient with single diagnosis ($p = 0.05$). There was also more prevalence of *Streptococcus pyogenes* and *Klebsiella pneumoniae* in patients with multiple diagnoses but with no statistical significance.

The prevalence of pathogenic bacteria in the different age groups of the patients was shown in Figure 3. The prevalence of *S. pneumoniae* was the highest in the younger age group and the prevalence decreased as the age increased.
**M. catarrhalis** was found most commonly in patients between 3 and 7 years old. **H. influenzae** had similar prevalence in all age groups with no statistical difference. **S. aureus** had ascending prevalence from the youngest group to the oldest group.

Univariate analysis of the type of bacteria and the patient’s age, adenoid size, and the diagnosis was in Table 3. The patient’s age had significant associations with the prevalence of **S. pneumoniae**, **M. catarrhalis**, and **S. aureus**. The prevalence of bacteria had no significant association with the size of the adenoid or the diagnosis of adenoid-related diseases.

Multivariate analysis between the effect of age, adenoid size, and diagnosis upon the outcome of the bacterial culture was in Table 4. Age was the only significant variable for **S. pneumoniae**, **M. catarrhalis**, and **S. aureus** when adjusted for the diagnosis and the size of the adenoid.

**Discussion**

Bacterial infection is one of the causes of adenoid hypertrophy and chronic infection of the adenoid.12 Bacterial infections of the adenoid are mostly polymicrobial, and they are the same pathogens that cause sinusitis and otitis media.6 From previous literature, potential pathogens identified in the adenoid were **S. pneumoniae**, **H. influenzae**, beta-hemolytic streptococci, **S. aureus**, and **K. pneumoniae**.13 There were significant associations of bacteria on the adenoid surface, in the adenoid core, and in the middle meatus of the patients with infection of the adenoid, sinusitis, or otitis media.14 These findings supported the concept that adenoid is the reservoir of the adenoid-related infection. The most common presentation of adenoidal diseases in this study was OSDB from adenoid hypertrophy. **H. influenzae** was the most common bacterial pathogen found in patients with OSDB, followed by **S. aureus** and **S. pneumoniae**. Our findings were similar to the study of Brodsky and Koch8 that **H. influenzae** was the most common agent found in the diseased adenoid comparing to the controls. Adenoid hypertrophy was associated with intracellular pathogens, including **S. aureus**, **S. pyogenes**, and **H. influenzae**.6 In the
In this study, *H. influenzae* was the most common organism found in both OME and sinusitis groups. Saafan et al.\(^{16}\) compared the bacteriology of the adenoid with OME and the adenoid with OSA using polymerase chain reaction (PCR) and scanning electron microscopy for biofilm, and they found that *H. influenzae* was the most common bacteria found in both groups, but the group with OME had a higher grade of biofilm formation. In this study, there was no statistical difference between the prevalence of bacteria in OSDB, OME, and sinusitis. All three groups had a similar pattern of prevalence. Brook et al.\(^{17}\) concluded that the distribution of organisms did not vary among the three groups of the

| Parameter | Number of patients | Number of bacteria (%) | S. pneumoniae | H. influenzae | M. catarrhalis | S. aureus |
|-----------|------------------|------------------------|---------------|---------------|---------------|-----------|
| Age       |                  |                        |               |               |               |           |
| 0–3       | 14               | 9 (64.3)               | 5 (35.7)      | 1 (7.1)       | 4 (28.6)      |           |
| >3–7      | 248              | 98 (39.5)              | 123 (49.6)    | 67 (27.2)     | 94 (37.9)     |           |
| >7–10     | 103              | 25 (24.3)              | 47 (45.6)     | 18 (17.5)     | 55 (53.4)     |           |
| >10       | 42               | 6 (14.3)               | 19 (45.2)     | 5 (11.9)      | 26 (61.9)     |           |
| p-value   | <0.001*          | 0.70                   | 0.03*         | 0.003*        |               |           |
| ANR       |                  |                        |               |               |               |           |
| 0–70      | 89               | 28 (31.5)              | 40 (44.9)     | 17 (19.1)     | 40 (44.9)     |           |
| >70–90    | 253              | 87 (34.4)              | 129 (51)      | 63 (24.9)     | 112 (44.3)    |           |
| >90       | 65               | 23 (35.4)              | 25 (38.5)     | 11 (16.9)     | 27 (41.5)     |           |
| p-value   | 0.85             | 0.17                   | 0.27          | 0.90          |               |           |
| Diagnosis |                  |                        |               |               |               |           |
| OME       | 306              | 105 (34.3)             | 149 (48.7)    | 69 (22.5)     | 136 (44.4)    |           |
| Sinusitis | 23               | 4 (17.4)               | 8 (34.8)      | 5 (21.7)      | 7 (30.4)      |           |
| OME       | 78               | 29 (37.2)              | 37 (47.4)     | 17 (21.8)     | 36 (46.1)     |           |
| p-value   | 0.20             | 0.44                   | 0.99          | 0.39          |               |           |

ANR: adenoid–nasopharyngeal ratio; OME: otitis media with effusion; OSDB: obstructive sleep-disordered breathing.

*Statistically significant.

| Parameter | S. pneumoniae | H. influenzae | M. catarrhalis | S. aureus |
|-----------|---------------|---------------|----------------|-----------|
| Age       |               | p-value       | p-value        | p-value   | p-value |
| 0–7       | 4.02          | 0.003*        | 1.14           | 0.693     | 2.63    | 0.053*   | 0.35     | 0.003*   |
| 7.1–10    | 1.85          | 0.217         | 0.97           | 0.938     | 1.53    | 0.439    | 0.68     | 0.309    |
| >10       | 1             | –             | 1              | –         | 1       | –        | 1        | –        |
| ANR       |               | p-value       | p-value        | p-value   | p-value |
| 0–70      | 1             | –             | 1              | –         | 1       | –        | 1        | –        |
| 71–90     | 1.13          | 0.656         | 1.31           | 0.280     | 1.37    | 0.309    | 1.04     | 0.883    |
| >90       | 1.06          | 0.871         | 0.77           | 0.435     | 0.78    | 0.563    | 1.01     | 0.971    |
| Diagnosis |               | p-value       | p-value        | p-value   | p-value |
| OME       | 1             | –             | 1              | –         | 1       | –        | 1        | –        |
| Sinusitis | 0.37          | 0.098         | 0.58           | 0.265     | 1.03    | 0.959    | 0.46     | 0.135    |
| OSDB      | 0.87          | 0.591         | 1.05           | 0.860     | 1.04    | 0.910    | 0.95     | 0.845    |

ANR: adenoid–nasopharyngeal ratio; OME: otitis media with effusion; OSDB: obstructive sleep-disordered breathing.

*Statistically significant.

Table 3. Univariate analysis between adenoid bacteria and age, adenoid size, and diagnosis.

Table 4. Multivariate analysis between adenoid bacteria and age, adenoid size, and diagnosis.
diagnosis (otitis media, adenotonsillitis, and obstructive sleep apnea), but the concentration of pathogenic bacteria per gram of the adenoid was higher in the diseased adenoid than in the control group. Antibiotics should be useful to decrease bacterial load in the diseased adenoid, as demonstrated in the study of Brook and Foote\(^\text{18}\) that the number of organisms in the diseased adenoid was reduced after the treatment with amoxicillin or cefprozil comparing to the controls.

Size of the adenoid is not considered when the patient has an indication for adenoidectomy in chronic sinusitis. The limitation of this study is the information regarding the size of the adenoid was retrieved from the measurement of the lateral skull X-ray without endoscopic findings. Flexible endoscopy is now becoming more popular for the evaluation of the adenoid as it can be performed in the outpatient setting. Adenoid–choana ratio can be measured from endoscopy with the picture recorded instantly without risking the child to the radiation exposure of the skull X-ray.

Age was associated with adenoid bacteriological findings in this study. \textit{S. pneumoniae} was more prevalent in young children under 7 years. \textit{H. influenzae} was found equally common in all age groups of the children in this study with high prevalence. These findings imply that \textit{H. influenzae} vaccine may be of great use with high impact on adenoid-related upper respiratory tract infections. Jeong et al.\(^\text{19}\) described the bacteriology of the tonsillar core culture according to age and \textit{S. pneumoniae} was more prevalent in patients younger than 8 years, whereas \textit{S. aureus} was more prevalent in older patients and \textit{H. influenzae} was found more commonly regardless of age.\(^\text{19}\) From the multivariate analysis, age was the only significant variable that had an association with types of bacteria in the adenoid. Bacteriology of the adenoid and the association with age should be useful for the selection of antimicrobials and prevention of adenoidal diseases with the specific bacterial vaccine.

**Conclusion**

Prevalence of pathogenic bacteria in the adenoid was similar in children with OSDB, OME, and chronic sinusitis. The most common bacterial agent found in the culture was \textit{H. influenzae}. Age had a significant association with the outcome of bacterial culture in univariate and multivariate analyses. Size of the adenoid and types of adenoid-related diseases had no significant association with the outcome of bacterial culture.

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**Declaration of conflicting interests**

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

**Ethical approval**

Ethical approval for this study was obtained from the Siriraj Institutional Review Board, Faculty of Medicine Siriraj Hospital, Mahidol University, Bangkok, Thailand (Certificate of Approval COA no. Si 070/2018).

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**Informed consent**

Informed consent was waived by the Institutional Review Board due to the nature of the retrospective study without identifiable data (SIRB Protocol no. 034/2561(EC2)).

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