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### **Original Article**

## Oral pathogens in children with respiratory disease



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#### ABSTRACT

*Objective*: Respiratory disease occurs frequently in children. The nasal cavity, trachea, and oral cavity are considered the primary routes of infection. We aimed to investigate the types of microbial pathogens present in the oral cavities of children with respiratory disease. In addition, we compared the detection rates of different pathogens between the oral cavity and the nasopharynx according to Hellman dental age.

Materials and methods: We included 32 children who were hospitalized for respiratory disease and classified them according to Hellman dental age. Furthermore, 32 control children were included as controls. Specimens were collected using two palate swabs: one taken at hospital admission and the other at discharge. The culture results of the samples were compared with those of the nasopharyngeal cultures.

Results: The following six bacterial strains were detected in both the palate and the nasopharynx: α-Streptococcus, Corynebacterium, Haemophilus influenzae, Haemophilus parainfluenzae, methicillin-resistant Staphylococcus aureus (MRSA), and Neisseria.

In the comparison of the detection rates, Neisseria were significantly more frequently detected in the palate. MRSA was detected at a significantly higher rate in the naso-pharynx. When the detection rates of different bacteria were compared according to Hellman dental age, significant differences were observed in the detection rates of  $\alpha$ -Streptococcus in stage IA and Neisseria in stages IC and IIA.

Discussion: We noted that the bacterial load of specific bacteria in the palate increased with Hellman dental age. Furthermore, the increase in bacterial load in the oral cavity at stage IA, when teeth have not yet erupted, suggests the need for oral management.

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#### 1. Introduction

In Japan, childhood mortality due to pneumonia and acute bronchitis has decreased over time. In 1950, the childhood mortality rates for pneumonia and acute bronchitis were 17.0% and 5.0%, respectively, whereas in 1990, these rates declined to 2.4% and 0.2%, respectively. The childhood mortality rate for pneumonia had further declined to 1.6%, and that for acute bronchitis had remained at 0.2% in 2011 [1]. However, despite the large overall decline in mortality due to respiratory diseases, they continue to occur relatively frequently in Japanese children [2–5]. Respiratory infections are particularly common in neonates and infants who have little immunity. In such cases, the nasal cavity and the trachea are considered as the primary routes of infection. Bacteria that colonize the nasopharynx are known to have similar characteristics to pathogens that cause respiratory tract infections [6-8]. Japanese health-care guidelines therefore stipulate the identification of pathogenic bacteria through sputum or nasopharyngeal cultures and the administration of effective antibacterial agents [9]. However, although the oral cavity is also considered as an important route of infection for respiratory infections, very few currently published studies have been conducted to compare the microbial pathogens present in the oral and nasal cavities.

The oral cavity is the entrance to the respiratory tract and is thus considered a route of entry for respiratory disease virulence factors. A relationship between oral bacteria and respiratory disease has also been suggested, of which ventilator-associated pneumonia is considered a typical example [10-13]. Colonization of the oral cavity by microbial pathogens has been reported to cause disease onset; studies have also reported the influx of oral secretion below the glottis through the outer surface of the tracheal intubation tube and similar observations [14]. Methods for preventing such occurrences have been examined from many perspectives, with oral care being considered as an important preventive measure [15-19]. Recent studies have also indicated the efficacy of oral care for preventing respiratory infections such as pneumonia and bronchitis [20-22]. Awareness of the importance of oral care in preventing respiratory infections in patients in acute care is increasing [23,24]. However, few similar studies have been conducted to assess the importance of oral cavity pathogens in respiratory infections in children.

The aim of the present study was to assess oral care methods in children with respiratory disease by examining the relationship between nasopharyngeal bacterial flora and the types and numbers of oral microbial pathogens in children with respiratory disease.

#### 2. Materials and methods

#### 2.1. Subjects

Thirty-two children (22 boys and 10 girls; hereinafter referred to as the "respiratory disease group") who were admitted to the pediatric ward of the Showa University Hospital for respiratory disease between July and November 2013 were recruited to the study. The age of the children in the respiratory disease group ranged from 1 month to 6 years 1 month, with a mean age of 1 year 10 months. We also included 32 control children (22 boys and 10 girls; hereinafter referred to as the "control group") who underwent oral health care at the Department of Pediatric Dentistry of the Showa University Dental Hospital. The ages of the children in the control group ranged from 1 month to 6 years, with a mean age of 1 year 10 months. An overview of the subjects' characteristics is shown in Table 1.

The respiratory disease group was classified according to Hellman dental age as follows: Hellman stage IA (predental period), eight children (mean age, 3 months); Hellman stage IC (primary tooth eruption), 16 children (1 year 6 months); and Hellman stage IIA (completion of primary dentition), eight children (4 years 3 months). The control group was classified according to Hellman dental stage as follows: Hellman stage IA, eight children (mean age, 3 months); Hellman stage IC, 16 children (1 year 6 months); and Hellman stage IIA, eight children (4 years 3 months).

The respiratory diseases under investigation were pneumonia, bronchial asthma, bronchiolitis, bronchitis, respiratory syncytial virus infection with respiratory symptoms, and asthma. Children with congenital diseases or underlying diseases were excluded from the study. Many children with respiratory diseases have multiple concurrent symptoms, and those with only one symptom are rare. Such cases are frequently given a single diagnosis; however, in most cases, the respiratory symptoms are misdiagnosed. Therefore, making an accurate definitive diagnosis is difficult. For example, cases with asthma symptoms have been diagnosed as pneumonia. Wheezing in children is caused by anatomical and physiological characteristics. The duration of hospitalization ranged from 3 to 14 days, with a mean of 6.8 days.

The guardians or legal representatives of all the children received an explanation of the study and provided written consent for their children's participation in the study.

#### 2.2. Specimen collection and culture methods

Specimens were collected using a sterile cotton swab (Seed Swab<sup>®</sup>y1, Eiken Chemical Co., Ltd., Tokyo, Japan). The subjects' deep palates were scraped with a dedicated sterile swab under fixed pressure for approximately 10 s; these swabs were then used as specimens [25,26]. After collection, the specimens were cultured in a laboratory at SRL, Inc. Cultures were prepared using the following media: blood agar, chocolate agar, deoxycholate hydrogen sulfide lactose agar, CHROMagar methicillin-resistant Staphylococcus aureus (MRSA), CHROMagar Candida, and phenylethyl alcohol agar. Cultures on chocolate agar were incubated in carbon dioxide and grown anaerobically at 35  $\pm$  2 °C, while cultures on all the other media were grown aerobically at  $35 \pm 2$  °C. After seeding the specimens on the culture media, bacteria were extracted after 48 h of culture, and simple determination was performed over a period of up to a maximum of 7 days. The method of identification was performed by Gram staining of colonies. From the results of Gram staining and colony characteristics, the colonies were classified into five groups: gram-positive cocci,

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