



## Current Issues in Pharmacy and Medical Sciences

Formerly ANNALES UNIVERSITATIS MARIAE CURIE-SKŁODOWSKA, SECTIO DDD, PHARMACIA

journal homepage: <http://www.curipms.umlub.pl/>



# Upper respiratory tract microbiota in health and disease – a minireview

EDYTA CHWIEJCZAK\*, URSZULA KOSIKOWSKA

Department of Pharmaceutical Microbiology with Laboratory for Microbiological Diagnostics, Medical University of Lublin, Chodzki 1, 20-093 Lublin, Poland

### ARTICLE INFO

Received 28 April 2018  
Accepted 17 May 2018

#### Keywords:

respiratory microbiota,  
pathology,  
infections.

### ABSTRACT

A most common infection is that the upper respiratory tract. The human body is inhabited by millions of microorganisms, most of which enable the proper functioning of some systems and constitute a barrier protecting against harmful external factors. Due to the continuous contact of the upper airway microbiota with the external environment, it may seem to be extremely unstable and very diverse. A number of studies have been conducted that have assessed the effect of various factors on the composition of the upper respiratory tract microbiota. This paper presents a literature analysis which showed that the anatomical area is rich in terms of the microbiota that forms it, and despite the constant changes in many parameters, including physical – very stable.

### INTRODUCTION

Symbiotic, as well as pathogenic bacteria are an integral part of the human body. Together they create the natural microflora of the body, recently referred to as microbiota [1,2]. The human microbiome forms about 10-100 trillion microorganisms, most of which belong to intestinal microbiota [3,4]. The airways microbiota is not as well explored as the microbiota of other body parts, e.g. skin or digestive system.

The year 2008 was the turning point for a deeper understanding of the human microbiota, when the Human Microbiome Project (HMP) was created by the National Institutes of Health (NIH) [4]. The aim of this project was to comprehensively assess the microbiome and determine its role in health and in the course of various diseases. In studies conducted as part of the HMP project, microorganisms isolated from the urogenital tract, gastrointestinal tract, skin, mouth and nasal cavity were evaluated. Due to the fact that traditional methods used in microbiological diagnostics were often unable to faithfully reproduce the micro-environment of the human body, they could cause trouble in proper identification, which is why the HMP assumed the study of microorganisms using molecular methods.

Human body and bacteria interact with each other. In health, when the host's immune system is functioning properly, the microorganisms that exist in its body are harmless and even necessary to maintain the state of

homeostasis [4]. In case of immunosuppression, imbalance in the composition of microbiota of mucous membranes or skin may lead to the development of opportunistic infections. Numerous studies have shown that the change in the human microbiota is closely related to the development of many illnesses – this fact suggests that manipulation of the human microbiota can be effective in the prevention and treatment of several diseases. Various experiments conducted with the use of probiotic microorganisms confirm that regulating gastrointestinal microbiota impacts the human health.

### MICROBIOTA OF THE RESPIRATORY SYSTEM

The development of the airways microbiota depends on many factors, including the availability of micro- and macro- elements, pH, temperature or the presence of oxygen, and many others [2,5]. Therefore, in the studies on human respiratory microbiota, information about the quality of the patient's respiration is necessary [4,6]. This is important not only due to the differences in partial gas pressures, but also due to the fact that it determines the migration of microorganisms inhabiting the mucous membranes of the human airways. Therefore, in conditions associated with respiratory disorders, the composition of the upper respiratory tract microbiota may change.

The natural nasopharynx microbiota of healthy adult humans include not only commensal microorganisms, but also, in many cases, pathogenic microorganisms such as hemophilic bacteria (*Haemophilus influenzae*),

\* Corresponding author

e-mail: edytachwiejczak@interia.pl

Gram-positive cocci (*Streptococcus pneumoniae*, *Staphylococcus aureus*) and Gram-negative cocci (*Neisseria meningitidis* and *Moraxella catarrhalis*) [7,8].

### NASOPHARYNGEAL MICROBIOTA IN ADULTS INFECTED WITH RHINOVIRUSES

Allen *et al.* [7] evaluated whether and how the rhinovirus infection affects the state of nasopharyngeal microbiota. In the group of volunteers who applied for the study, the nasopharynx microbiota was evaluated before, during and after viral infection. Microbes belonging to three phyla: *Firmicutes*, *Actinobacteria* and *Proteobacteria* were the most frequently isolated from the nasopharynx of examined patients. However, differences in the occurrence of individual microorganisms were found at the level of genera. An important difference was determined in the presence of bacteria of the genus *Neisseria* spp. and *Propionibacterium* spp. between individuals infected and uninfected with a virus. These studies also found that the occurrence of *Haemophilus* spp. in infected samples was much lower compared to the occurrence of these bacteria in healthy patients.

Next, the researchers compared the composition of upper respiratory microbiota with microorganisms inhabiting other areas of the human body, including skin, hair, ears, tonsils and the digestive system. This analysis revealed that bacteria inhabiting a specific region of the body form a single, logical cluster in which similar species of microorganisms predominate. This is most likely caused by the close setting of these areas, which allows continuous migration of microorganisms.

### UPPER RESPIRATORY MICROBIOTA IN INFLUENZA AH1N1 VIRUS INFECTION

Chaban and colleagues [9] have shown that the microbiota of the upper respiratory tract, and especially the nasopharynx, is dominated by three phyla of microorganisms: *Actinobacteria*, *Firmicutes* and *Proteobacteria* (Fig 1.). The samples tested were from patients who had been infected with the AH1N1 influenza virus during the pandemic in 2009. Patients were of different ages, races and sex. The material was gathered in the hospital, as well as from community patients. The identification of microorganisms was based on molecular biology methods using quantitative PCR (qPCR), which enabled the detection of characteristic sequences in the genome of the microorganisms studied. Studies have shown that a large proportion of patients (27/67, 40% of study subjects) were colonized mainly with bacteria belonging to the *Varovorax paradoxus* species (belonging to the *Proteobacteriae* phylum) and *Enterococcus hirae* (phylum *Firmicutes*). Phylogenetic analysis showed that the most often occurring microorganisms in the nasopharynx of the examined patients were: *Firmicutes* (42.5%), *Proteobacteria* (27.7%) and *Actinobacteria* (21.7%). Presence of other microorganisms, including *Bacteroides* and fungi was also indicated in the diagnostic samples. However, they occurred in less significant amounts than the bacterial types listed above.

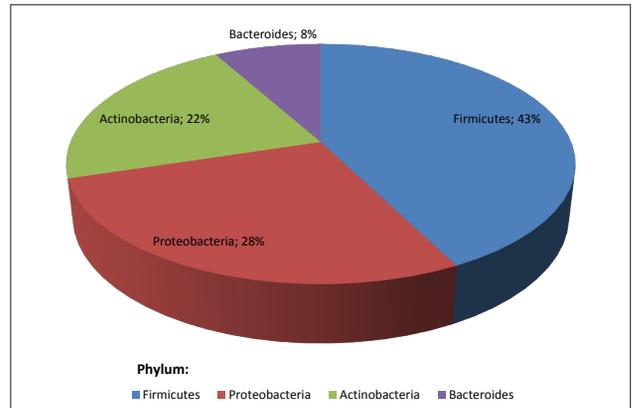


Figure 1. Diversity of bacterial phyla in the nasopharyngeal microbiota of the patients

Researchers showed that among patients infected with the AH1N1 influenza virus, the upper respiratory tract microbiota composition is not dependent on sex, general health or the place of sample collection [9]. The same three phyla of microbes were always dominating. It has also been shown that the richness and diversity of microorganisms that form the nasopharyngeal microbiota increases with older age among the patients infected with AH1N1.

### NASOPHARYNGEAL MICROBIOTA IN PATHOLOGICAL CONDITIONS

A team of researchers from Texas decided to determine changes in the infant's nasopharyngeal microbiota during viral infection of the upper respiratory tract and during acute otitis [10]. These diseases most often occur in infants, and ear infection is usually a complication of the viral upper respiratory tract infection [11].

The studies evaluated the composition of nasopharyngeal microbiota in children in the first year of life [10]. Samples were collected every month up to 6 months and then at 9 months of age and during viral infection of the upper respiratory tract or during acute otitis. The identification of microorganisms was based on molecular methods. Four phyla of microbes were dominating in the isolates tested: *Proteobacteria* (39%), *Firmicutes* (26%), *Actinobacteria* (26%) and *Bacteroides* (6%). It was shown that the most common occurring in the microbiota of nasopharyngeal infants among the commensal organisms were *Actinobacter* spp., *Corynebacterium* spp. and *Dolosigranulum* spp. The dominant genera of pathogenic bacteria were *Pseudomonas* spp. and *Staphylococcus* spp. Moreover, the infant's nasopharyngeal were found to be colonized by pathogenic bacteria such as: unencapsulated *H. influenzae* (NTHi – Nontypical *H. influenzae*) *S. pneumoniae* and *M. catarrhalis* early in life [12].

Researchers also analyzed the composition of nasopharyngeal microbiota with regard to antibiotic data. The most commonly used antibiotic was amoxicillin. It was shown that taking this drug did not significantly affect changes in the incidence of pathogenic microorganisms but significantly reduced the abundance of *Corynebacterium*, *Enterobacter*, *Staphylococcus* and *Dolosigranulum* species, while increasing the incidence of phylum *Firmicutes* in this type of *Bifidobacterium* spp.

Literature data shows that viral infections that often occur in children can significantly alter the composition of the upper respiratory tract microbiota. This is most often caused by the effect on microbial adhesion and the ability to colonize. The impact of viral infections on the general condition of the host's immune system is also significant and may result in changes in the composition of the upper respiratory tract microbiota [7].

### INFLUENCE OF THE DELIVERY TYPE ON THE UPPER RESPIRATORY TRACT MICROBIOTA IN CHILDREN

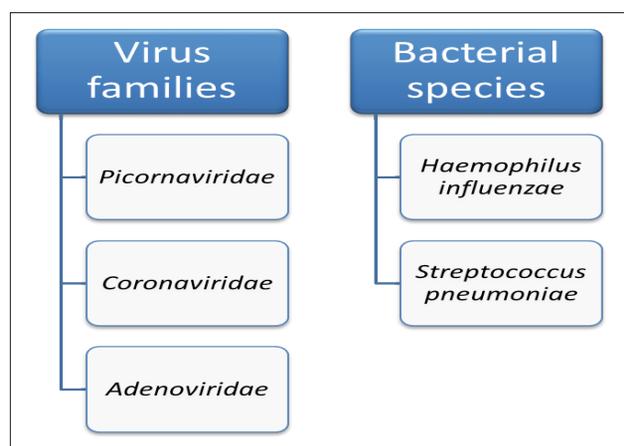
The human body is exposed to microbes in the first minutes of its life. The natural microbiota of the mother's body plays a key role in the health of the child – the microorganisms that form it will start to form the infant's microbiota during the delivery [13,14]. Currently, delivery by Caesarean section is a very common procedure. Statistically, the number of children who came into the world through the Caesarean surgery is much closer to the number of natural childbirths. During natural childbirth, the child "meets" the microorganisms that colonize the genital tract and the digestive system of the mother, while in the case of the Caesarean section, with the bacteria present on the skin and in the environment [13].

In a study conducted by Bosch *et al.* [13], it was assessed whether there is a difference in the occurrence of microorganisms that inhabit the nasopharynx of newborns which have been born naturally or through a Caesarean section. The predominant microorganisms colonizing the upper respiratory tract in children during the first 6 months of life were respectively: *Corynebacterium* spp. (25%), *Moraxella* spp. (21%), *Staphylococcus* spp. (19%) and *Streptococcus* spp. (11%). Immediately after the birth the microflora is very similar in all children. These are mainly microorganisms that colonize the mother's reproductive tract, her digestive system and skin, but also the microorganisms present in the environment – it is independent of the type of birth. Interestingly, the researchers showed that in a very short time (about the first week of life) the nasopharynx is colonized by the *S. aureus*. However, it is also rapidly displaced by *Corynebacterium* spp., then *M. catarrhalis* and *M. nonliquefaciens*, *S. pneumoniae* or *H. influenzae*. All these dynamic changes in the composition of the natural flora of the upper respiratory tract in children occur only within the first six months of life. Early colonization with *S. aureus* has a beneficial effect on the child's body, because it facilitates colonization of the upper respiratory tract by commensal strains like *Corynebacterium* spp. or *Moraxella* spp., which stabilize the ecological niche and reduce the risk of infection. Researchers have shown that early colonization of the upper respiratory tract with microorganisms belonging to the *H. influenzae* species causes greater susceptibility for mild and chronic infections such as bronchitis, lungs or middle ear diseases. Based on the presented results, it was found that the type of delivery has a moderate, direct impact on the nasopharyngeal microbiota in children. It has been shown that in infants delivered via Caesarean section microbiota during the first six months of life was less stable and much less diverse in terms of the presence of commensal species beneficial

to the respiratory system, such as *Corynebacterium* spp. or *Dolosigranulum* spp.

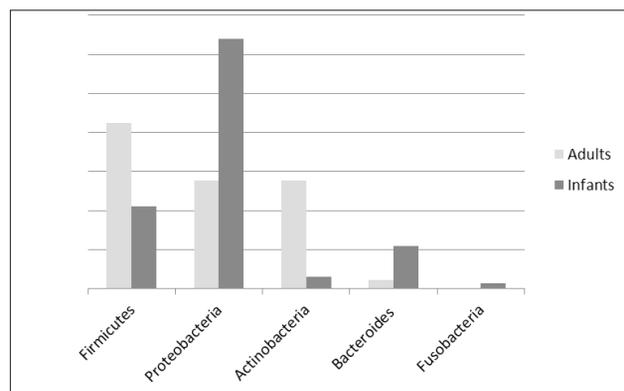
### VARIETY AND VARIABILITY OF NASOPHARYNX MICROBIOTA IN CHILDREN

One of the most common causes of disease in children is upper respiratory tract infections. These types of infections can have substantial consequences and even lead to patient's death. Typically, these types of infections have a viral background; bacterial infections occur less frequently [15]. The most common etiological factors of bacterial infections are pathogenic microorganisms such as *H. influenzae*, *S. pneumoniae*, *S. aureus* [16] (Fig. 2.). These microorganisms are often part of the natural microbiota of the upper respiratory tract and their carriers are asymptomatic – they are dangerous only in states of reduced immunity.



**Figure 2.** The most common etiological agents of upper respiratory tract infections in children [15]

Bogaert *et al.* [8] evaluated the nasopharyngeal microbiota among healthy one-and-a-half year old children. The analysis of the obtained data shows that five phyla of microorganisms prevailed among the samples tested: *Proteobacteria* (64%), *Firmicutes* (21%), *Bacteroides* (11%), *Actinobacteria* (3%) and *Fusobacteria* (1.4%). The most common genera were: *Moraxella* spp., *Haemophilus* spp., *Streptococcus* spp. and *Flavobacterium* spp. This fact suggests that the frequency of specific bacteria occurrence is dependent on age (Fig. 3). Researchers also assessed which microorganisms prevailed in the mouth and which in the nostrils.



**Figure 3.** Comparison of specific phyla prevalence in nasopharyngeal microbiota of healthy infants and adults [7,8]

It was shown that bacteria classified into the phyla *Firmicutes*, *Proteobacteria* and *Bacteroides* most often appeared in the mouth, while *Actinobacteria* and *Firmicutes* phyla were mainly present in the nostrils. This comparison shows that the dynamics of changes in the nasopharyngeal microorganisms is very diverse.

The carried out research showed that in samples taken in late autumn and winter, microorganisms belonging to the *Proteobacteria*, *Fusobacteria* and *Cyanobacteria* phylum were more frequent and abundant compared to samples collected in spring. However, the *Bacteroides* phylum appeared more often in the samples obtained in the spring. It was also noticed that the occurrence of such species as *Bacteroides* spp., *Bacillus* spp. or *Lactobacillus* spp. is extremely important for the general condition of airway microbiota and consequently – health. These microorganisms, by way of the production of bacteriocin, protect against pathogenic microorganisms to which the body is exposed, especially during autumn or winter. The increased abundance of these „beneficial” bacteria in human respiratory microbiota in spring may suggest a more balanced status of the upper respiratory tract microbiota and thus a better natural protective barrier of the body.

## CONCLUSION

In conclusion, a literature analysis of nasopharyngeal microbiota shows that this small region of our body can be inhabited by many different groups of microorganisms. They form a very rich niche, in which there are many dynamic changes. The microbiota of the upper respiratory tract has an extremely strong regenerative capacity because many infections with pathogenic microbes or viruses do not change the composition of microbiota in a sustainable manner – it could only lead to slight changes in the amount of these bacteria.

Despite many studies and analyzes of thousands of samples, natural upper respiratory tract microbiota and changes occurring within have not been fully understood and require further investigation.

## REFERENCES

1. Cui L et al. The Microbiome and the lung. *Annals ATS*. 2014;11: 227-32.
2. Ursell LK, Metcalf JL, Wegener Parfrey L, Knight R. Defining the human microbiome. *Nutr Rev*. 2012; 70 (Suppl 1): S38-S44.
3. Gorecka D, Nowinski A, Augustynowicz-Kopec E. Respiratory system microbiom (in polish). *Pneumonol Alergol*. 2014;82:481-5.
4. Malinowska M, Tokarz-Deptula B, Deptula W. Microbiome of the respiratory system under physiological and pathological conditions (in polish). *Post Mikrobiol*. 2016;55(3):279-83.
5. Andrzejczak A, Chwiejczak E, Kosikowska U, Stepien-Pysniak D, Malm A. Phenotypic diversity of *Haemophilus influenzae* and *Haemophilus parainfluenzae* isolates depending on origin and health condition. *Curr Issues Pharm Med Sci*. 2017;30:90-9.
6. Kosikowska U, Rybojad P, Stepien-Pysniak D, Zbikowska A, Malm A. Changes in the prevalence and biofilm formation of *Haemophilus influenzae* and *Haemophilus parainfluenzae* from the respiratory microbiota of patients with sarcoidosis. *BMC Infect Dis*. 2016;16:499.
7. Allen KE, Koeppl AF, Hendley OJ, Turner SD, Winther B, Sale MM. Characterization of the nasopharyngeal microbiota in health and during rhinovirus challenge. *Microbiome*. 2014;2:22.
8. Bogaert D et al. Variability and diversity of nasopharyngeal microbiota in children: A metagenomic analysis. *PLoS ONE*. 2011;6
9. Chaban B, Albert A, Links MG, Gardy J, Tang P, Hill JE. Characterization of the upper respiratory tract microbiomes of patients with pandemic H1N1 influenza. *PLoS ONE*. 2013;8(7):69559
10. Chonmaitree T et al. Nasopharyngeal microbiota in infants and changes during viral upper respiratory tract infection and acute otitis media. *PLoS ONE*. 2017;12:0180630.
11. Chonmaitree T et al. Viral upper respiratory tract infection and otitis media complication in young children. *Clin Inf Dis*. 2008;46:815-823
12. Robinson J. Colonization and infection of the respiratory tract: What do we know? *Paediatr Child Health*. 2004;9:21-4.
13. Bosch A et al. Development of upper respiratory tract microbiota in infancy is affected by mode of delivery. *EBioMed*. 2016;9:336-45
14. Dominguez-Bello MG et al. Delivery mode shapes the acquisition and structure of the initial microbiota across multiple body habitats in newborns. *PNAS*. 2010;107:11971-5.
15. Cotton MF, Innes S, Jaspan H, Madide A, Rabie H. Management of upper respiratory tract infections in children. *S Afr Fam Pract*. 2008;50:6-12.
16. Thapa S, et al. Burden of bacterial upper respiratory tract pathogens in school children of Nepal. *BMJ Open Resp Res*. 2017;0:000203.