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The efficacy and tolerability of Streptococcus salivarius 24SMB and Streptococcus oralis 89a administered as nasal spray in the treatment of recurrent upper respiratory tract infections in children

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Abstract. – OBJECTIVE: Nasal administration of *Streptococcus salivarius* 24SMB and *Streptococcus oralis* 89a has been proposed to reduce the risk of new episodes of adenoiditis, tonsillitis and acute rhinosinusitis in children.

PATIENTS AND METHODS: We enrolled 202 children with a recent diagnosis of recurrent upper respiratory tract infection. All the patients were treated twice daily for 7 days each month for 3 consecutive months with a nasal spray whose active agents were two specific bacterial strains: *Streptococcus salivarius* 24SMB and *Streptococcus oralis* 89a. Evaluation was performed at the end of treatment and at follow-up at 3, 6, and 12 months.

RESULTS: Patients who completed the entire 90-day course of bacteriotherapy and the follow-up period showed a 64.3% reduction in their episodes of upper respiratory tract infections compared to the number of episodes recorded in the previous year. Treatment decreased the reported incidence of infection events by 52.4% in the first 3 months, 31.2% at 6-month follow-up, and 20.8% after 12 months. Enrolled patients tolerated the product well, and there were no dropouts.

CONCLUSIONS: Prophylactic bacteriotherapy by administration of *Streptococcus salivarius* 24SMB and *Streptococcus oralis* 89a in children with a history of recurrent upper respiratory tract infection could reduce the number of episodes of otolaryngologic infections. Bacteriotherapy can be even more clinically important due to increasing difficulty in finding new effective antibiotic compounds. New alternative therapeutic approaches must be found with, in comparison to antibiotics, greater specificity and safety with respect to patients' native beneficial flora; lack of drug interactions; the ability to leverage complementary systemic modes of action; and drastically reduced risk of developing resistance within the patient population and the environment.

Key Words

Streptococcus salivarius 24SMB, *Streptococcus oralis* 89 a, Pharyngitis, Adenotonsillitis, Acute otitis media.

List of Abbreviations

URT: upper respiratory tract; TLR: Toll-like Receptors; COPD: Chronic Obstructive Pulmonary Disease; LPS: lipopolysaccharide; AOM: acute otitis media.

Introduction

The capability of microbes to adapt to their environment is evident in the human body as in other living organisms. Commensal microorganisms and their metabolites maintain the stability of their habitat and the health of their host as a symbiotic system. Even inflammation plays a role in the creation of a microenvironment in which certain microbes can or cannot grow and reproduce[1,2]. Consideration of such changes in the microbiome as cause or effect of the counteraction of the host defense is still debated, particularly in a number of chronic inflammatory conditions associated with disrupted epithelia. The occupation of upper respiratory tract (URT) niches by indigenous flora can induce resistance against other pathogens. As a result, an invading pathogen has the unenviable task of competing with established commensal flora for adhesion receptors and nutrients on the mucosal surface – a competition that limits the chances of adhesion, replication, a dissemination of disease.

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Microbes can physiologically develop the capacity to inhibit competing microbes using antimicrobial compounds. A prime example is the production of hydrogen peroxide by *Streptococcus pneumoniae*, which is bactericidal for *Staphylococcus aureus*[3]. Moreover, some bacterial species produce peptides with antimicrobial efficacy, generally referred to as bacteriocins[4]. This mechanism is clearly evident in the commensal streptococcal species *Streptococcus salivarius*, which inhibits pathogenic *Streptococcus pyogenes* growth by the production of bacteriocins. The large surface of the nasal cavity contains niches shaping the ecosystem's habitat, and antibiotic treatment as well as invading pathogens may break this balance[5]. Indigenous microbiotic species make a large contribution to resisting colonization by invading pathogens[6]. The presence of so-called keystone species is strongly related to the maintenance of biodiversity in ecological communities; their loss results in the extinction of many other (micro-) organisms, hence decreasing the community's biodiversity[7].

The diversity of the microbiota used by commensal bacteria and pathogens is mainly due to the mechanisms of molecules of the innate immunity[8-10], specifically the expression of different Toll-like Receptors (TLRs) on the respiratory epithelial cells. In this case, an important role is played by inflammation. Bacteria that induce low-grade asymptomatic inflammation may create conditions enabling easier penetration and invasion by pathogenic bacteria. For example, colonization with proteobacteria such as non-typeable *Haemophilus influenzae*, which is accompanied by mucosal inflammation, is characterized by increased epithelial thickness of the large airways, production of early pro-inflammatory cytokines, and damage of the epithelial barrier function[11].

In contrast, in Chronic Obstructive Pulmonary Disease (COPD), commensal strains such as Prevotella, antagonize pathogens lipopolysaccharide (LPS)-mediated TLR4-signalling with dampening and downstream signaling, contributing to mucosal homeostasis and colonization resistance *vs.* pathogens[12].

Skewed inflammatory mediator expression can also be influenced by environmental factors such as cigarette smoke[13]. These factors can also influence the interplay between innate immunity and mucosal microbial communities. The delicate balance of these factors makes the healthy microbiome an important component of the epithelial barrier[14].

Focusing now on the commensal microorganisms of the upper airways, the α-hemolytic streptococcus is mostly present in the nasopharynx and may interfere with survival and multiplication of pathogens that are most often associated with development of acute otitis media (AOM) and its consequences such as acquired hearing loss[15,16]. More recently, specific types of α-hemolytic streptococcus have been isolated from the pharynx of healthy subjects: *Streptococcus salivarius* 24SMB and *Streptococcus oralis* 89a. These microbes are potential nasopharyngeal probiotics, with immunomodulatory and anti-inflammatory skills, production of plasmin-encoded bacteriocins and a good safety profile [17-19]. The first study with *Streptococcus salivarius* 24SMB, recently published by Marchisio et al [20], reported significant activity against AOM pathogens when *Streptococcus salivarius* is administrated intranasal in otitis-prone children.

The aim of this study is to evaluate the role of *Streptococcus salivarius* 24SMB and *Streptococcus oralis* 89a in the treatment and prevention of recurrent URT infections in children.

Patients and methods

Two hundred and two patients presenting to our University Hospital from September 2016 to February 2017 were included in the study. The study was specifically approved by the Ethical Committee of our University Hospital. One-hundred forty-one subjects were males and 61 were females, with a median age of 7.5 years. The patients were included after the collection of anamnestic data and clinical evaluation that reported recurrent URT pathology treated with systemic antibiotic therapy by own family physician. The URT conditions analyzed in the present study were adenoiditis, tonsillitis, and acute rhinosinusitis.

Patients with severe concomitant diseases or lacking consent from their parents/caregivers were excluded from the study. All patients were treated with a nasal spray made up of a suspension of two specific bacterial strains: *Streptococcus salivarius* 24SMB and *Streptococcus oralis* 89a. This topical treatment was administered via intranasal spray using a disposable device twice daily for 7 days per month for 3 consecutive months. Patients were previously treated with an antibiotic and started the new intervention after 10 days of washout from antibiotic therapy. The treatment was concomitant with saline nasal irrigations in 40% of patients, with vasoconstrictor drops in 6% and with oral antihistamines in 6%. A questionnaire about the subjective efficacy of the therapy was collected from adult patients and from children's parents and/or caregivers and compared to clinical improvement of symptoms. Results obtained during therapy were compared with those of the previous year for each subject as reported by their parents.

Results

One year after the beginning of bacteriotherapy, a 64.3% of reduction in the number of observed events compared to the previous year was reported (Figure 1); similar results were observed for the number of recurrent episodes in specific URT pathologies.

The frequency of recurrences was evaluated as the reduction of episodes during follow-up at 3, 6, and 12 months. The highest percentage of URT events (57.3%) occurred in the first three months of treatment with a progressive decrease over the remaining period (24.9% at 6 months and 17.8% after 12 months). This overall trend was confirmed for each pathological condition: the recurrence percentage in patients affected by adenoiditis was 48.2% during the first 3 months, 29.5% at 6 months and 22.3% at the final follow-up visit; in patients affected by adenotonsillitis the recurrence rate was 51.7% during the first 3 months, 23.8% at 6 months and 24.5% at final follow-up; recurrence in patients affected by rhinosinusitis was 71.9% during the first 3 months, 21.3% at 6 months and 6.8% at final follow-up (Figure 2).

These results were confirmed by the subjective self-administered questionnaires on the efficacy of the therapeutic protocol: 58.7% of the patients reported a positive change in their symptoms and recurrence rate; 36.1% reported doubtful changes and improvement; 8% of treated patients reported a worsening of symptoms. Among symptoms collected from the questionnaires, a statistically significant improvement (*p* <0.05) was observed for nasal obstruction and ear fullness (Figure 3).

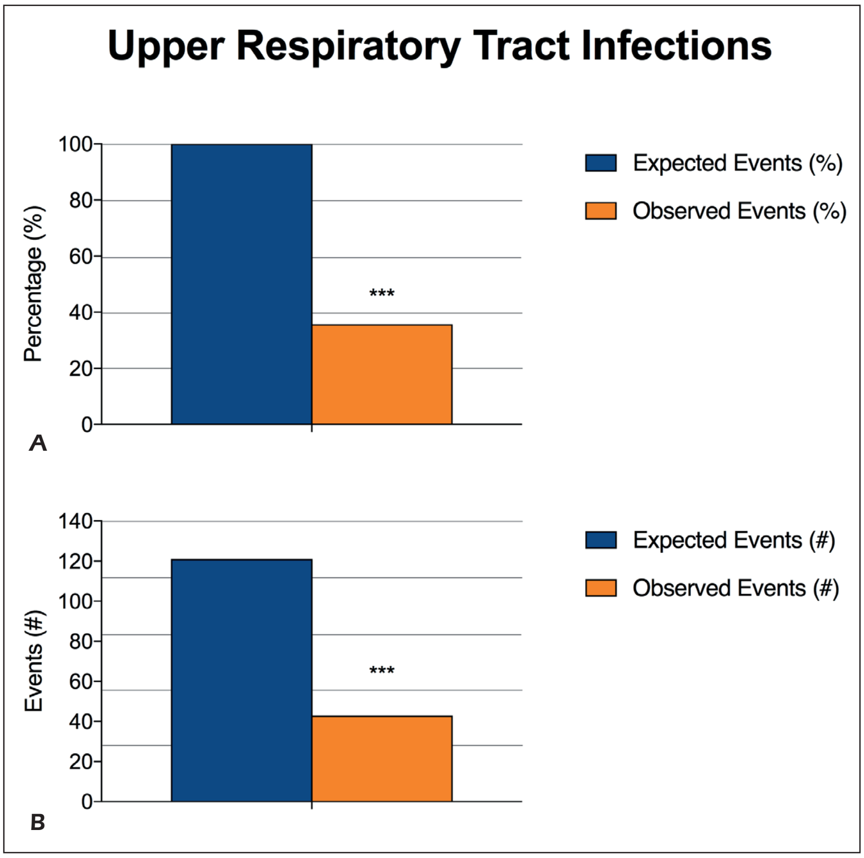

Figure 1. Data collected 12 months after beginning treatment. **A**, Percentage of upper respiratory tract infections in enrolled patients compared to data of the previous year from the same patients. **B**, Number of upper respiratory tract infections in enrolled patients compared to data of the previous year from the same patients.

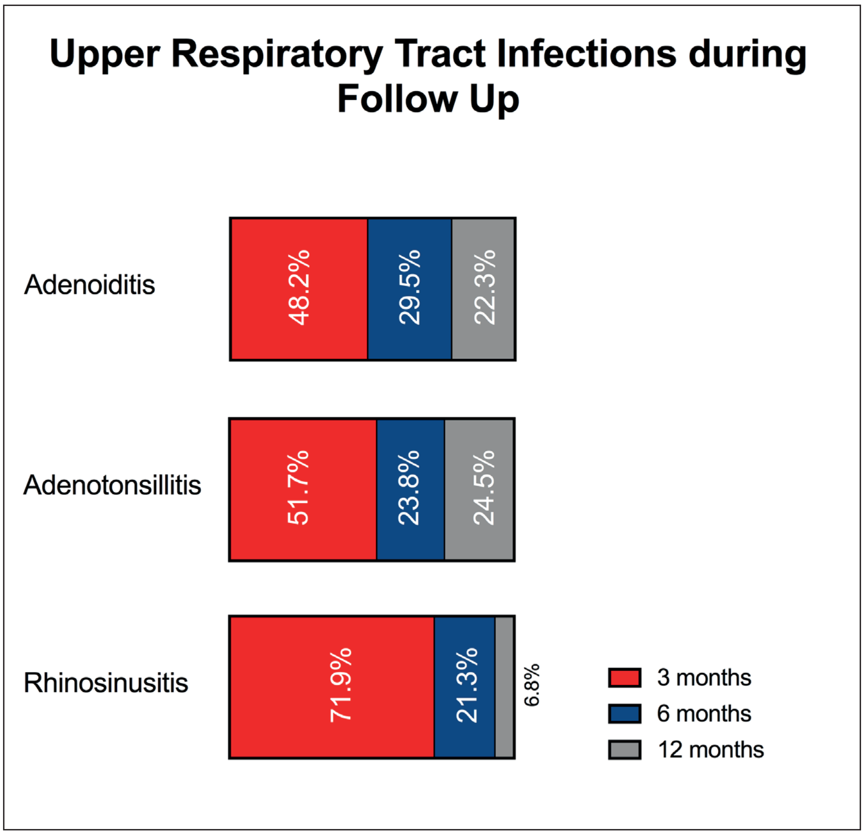

Figure 2. Data collected during follow up. Data collected during follow up 3, 6, and 12 months after the beginning of treatment for each pathological condition (adenoiditis, tonsillitis, rhinosinusitis).

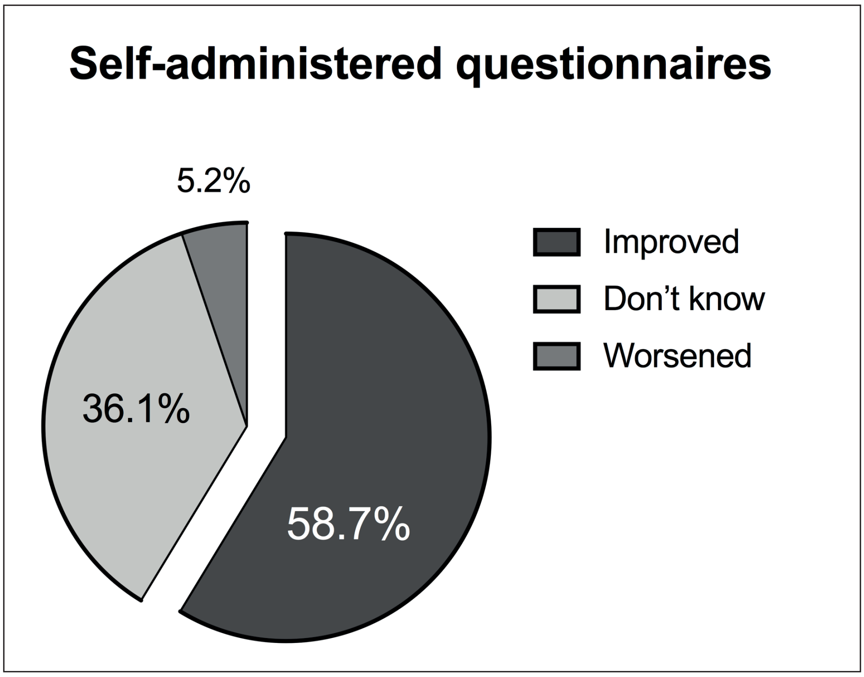

Figure 3. Subjective report on the efficacy of the therapeutic protocol. Subjective report on the efficacy of the therapeutic protocol recorded 12 months after the beginning of treatment using self-administered questionnaires. Most of the patients reported an improvement in symptoms; a small percentage reported a worsened condition.

Discussion

URT infections are the most common cause of consultation both for general practitioners and otolaryngologists worldwide, and they represent an important cause of reduced school days, work loss, and increased healthcare expenses. The significant burden of care in otolaryngologic pathology is represented by the need for medical examination, prescription of antipyretic medicaments, and often antibiotics. Beyond direct costs, URT infections involve high indirect costs in terms of loss of educational access and progress for the child kept home to rest, and economic productivity of the parental caregiver. Such negative effects are multiplied in the case of recurrent infection [21].

Unfortunately, inflammation of the rhinopharynx and oropharynx in children with adenotonsillar hypertrophy is underestimated and poorly treated, leading to complications in the middle ear and paranasal sinuses. Middle ear complications follow impaired ventilation following tubal obstruction. Viral or allergic inflammation of the nose leads to nasal congestion and osteomeatal complex obstruction with reduced ventilation of the paranasal sinuses. Such conditions, especially in asthmatic children [22,23], facilitate the ascent of respiratory pathogens (*Streptococcus pneumoniae*, *Haemophilus influenzae* and *Moraxella catharralis*) that colonize the nasopharynx, may spread to the tympanic cavity and paranasal sinuses, and may subsequently lead to complications in the middle ear and paranasal sinuses that mainly manifest later in adulthood.

Usually the treatment of these conditions requires topical or systemic anti-inflammatory drugs or antibiotics. On the other hand, excessive use of antibiotics, especially in Western countries, has resulted in the selection of strains resistant to treatment. Resistance becomes even more important in light of the increasing difficulty in finding new effective antibiotic compounds. Against this backdrop, alternative therapeutic approaches must be found that offer greater specificity and safety, avoid drug interactions, and leverage rather than compromise native complementary modes of action against microbial invaders, in order to improve health while drastically reducing the risk of further developing resistance. In the present study, we noted that follow-up at 12 months would approximate seasonal conditions at month zero, and thus suspect seasonal influence on symptoms to be minimal compared to therapeutic effect. Therefore, we attribute the correlation of increased amelioration with longer exposure to therapy to the need for sufficient intervention to obtain a satisfactory colonization of the URT.

Conclusions

Treatment and prevention of otolaryngologic diseases in children are now a primary focus of pediatric medical care. The topical application of streptococcal probiotics is a relatively undeveloped field but is becoming an attractive approach for both prevention and therapy, especially for pediatric age patients[24]. In this context, our data confirm the efficacy of *Streptococcus salivarius* 24SMB and *Streptococcus oralis* 89a as suitable bacteriotherapeutic agents against URT infections.

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Conflict of Interests

The authors declare that they have no conflict of interest.

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