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PRODUCTION OF BACTERIOCIN-LIKE INHIBITORY SUBSTANCES (BLIS) BY *STREPTOCOCCUS SALIVARIUS* STRAINS ISOLATED FROM THE TONGUE AND THROAT OF CHILDREN WITH AND WITHOUT SORE THROAT

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SHORT COMMUNICATION

ABSTRACT

Streptococcus salivarius strains, isolated from children with and without sore throat, were tested for bacteriocin production against *Streptococcus pyogenes. S. salivarius* strains producing bacteriocin-like inhibitory substances (BLIS) against *S. pyogenes* were more frequently found in children without sore throat. These results suggest that these children may be protected against sore throat by the presence of BLIS-positive *S. salivarius* strains.

Key-words: *Streptococcus salivarius, Streptococcus pyogenes*, bacteriocin-like-inhibitory-substances, BLIS

Bacterial interference has been recognized as a tool to prevent certain diseases. Several researchers believe that this is a natural protection against some pathogenic bacteria. *Streptococcus* are usually isolated from the mouth and are the dominant bacteria of this habitat. Among them, *S. salivarius,* a non pathogenic bacterium, is numerically the most significant of those on the tongue (5). Antagonism between *Streptococcus*, based on production of bacteriocins, has been investigated (2, 12).

The term "bacteriocin" was first coined by Jacob *et al.* (4). More recently, this term has been used to designate bacterial peptides and protein antibiotics which are ribossomally produced or derived from gene-encoded precursor peptides, and towards which the producing strain possesses a specific self-protection mechanism (8). Inhibitors that do not fulfill this criteria should be classified as bacteriocin-like inhibitory substances (BLIS) (12).

A wide range of interactions have been observed within oral bacteria including interactions with agents of throat infection. The potential protective role played by the resident microflora was explained by Sanders (11), who compared the inhibitory viridans streptococci in the throat of children infected with Group A streptococci with children who had not been infected. This inhibitory activity was present in children without *S. pyogenes* and was independent of the Group A test organism and inoculum size. Fantinato and Zelante (2) noted that among the oral bacteria which produce BLIS

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against *S. pyogenes, S. salivarius* was one of the most common in the mouth.

The aim of the present study was to verify if the presence of BLIS-positive *S. salivarius* strains in the mouth and throat of children with and without sore thoats, could be associated to protection against throat infection.

The samples were collected from 54 children who had frequently experienced sore throat (Group I) and from a control group of 50 children who had not experienced this disease in the last 3 years (Group II). The samples of Group I were collected in the Taubate Hospital (São Paulo - Brazil), from children with clinically diagnosed throat infections. The samples of Group II were collected from an Intermediate School in S. José dos Campos (São Paulo - Brazil), from children who did not experience sore throats. All children were 7 to 14 years old. A consent form and a questionaire about the frequency of the illness were full filled by the parents or guardians of all children.

Samples were collected by swabbing the tongue and throat. Mitis Salivarius Agar (Difco) was used to isolate *S. salivarius* strains. Ten colonies from each plate were submitted to biochemical identification, following the scheme outlined by Hardie and Bowden (3). Bacteriocin production was tested by performing the deferred antagonism test (1), using *S. pyogenes* ATCC 8133 (Type 23) as the indicator strain.

Any child was presenting BLIS positive *S.salivarius* strains either on the tongue or in the throat was considered a carrier.

Results in Table 1 show that 53.7% of children from Group I and 98% of children from Group II were BLIS-positive *S. salivarius* carriers. A statistically significant difference between results for Group I and II was found, at 5% level.

The results suggest that the presence of BLIS-positive *S. salivarius* strains in children who had not developed sore throats may be related to protection against throat infection.

As noted by Pichichero (6), sore throat is one of the most commom problems in children and Group A *Streptococcus* infections account for nearly all sore throats of bacterial etiology. The author also commented about the therapy failure in 20% of children under antibiotic treatment.

Sanders *et al.* (10) evaluated the effects of orally administered antibiotics on the composition of the normal throat flora and its ability to inhibit the growth of group A streptococci; the interfering activity of the flora was remarkedly diminished or absent during penicillin therapy.

Several studies were performed on the treatment of recurrent streptococcal tonsilitis by Roos *et al.* (7). Alpha-streptococci, inoculated into the throat of the patients, presented a successful protective effect against recurrence.

S. salivarius can be considered more appropriate than other streptococci for prevention of throat infections since this species is one of the most active antagonist against group A streptococci.

RESUMO

Produção de substâncias inibidoras semelhantes à bacteriocina por cepas de *Streptococcus salivarius,* isoladas da língua e garganta de crianças com e sem dor de garganta

Cepas de *Streptococcus salivarius,* isoladas de crianças com e sem dor de garganta, foram testadas quanto à produção de bacteriocina contra *Streptococcus pyogenes*. Os resultados mostraram que as crianças que não tinham dor de garganta

Table 1 - Isolation of BLIS[a] producing *Streptococcus salivarius* strains from children with and without sore throats

Group[b]	Number of Children	Total of strains[c]		Total of BLIS + strains		BLIS + strains Carriers
		Tongue	Throat	Tongue	Throat	
I	54	46/85.1	48/88.8	24/44.4	22/40.7	29/53.7
II	50	48/96.0	49/98.0	43/86.0	43/86.0	49/98.0
Total	104	94/90.3	97/93.2	67/64.4	65/62.5	78/75.0

[a] = Bacteriocin-like inhibitory substances
[b] = Group I, children with sore throat
Group II, children without sore throat
[c] = Number/%

possuiam, na boca, cepas de bactérias produtoras de substâncias inibidoras semelhantes à bacteriocina contra *S. pyogenes*.

Palavras-chave: *Streptococcus salivarius, Streptococcus pyogenes*, substâncias inibidoras semelhantes à bacteriocina, BLIS

REFERENCES

1. Crowe C C, Sanders W E Jr, Longley S. Bacterial interference. II. Role of the normal throat flora in prevention of colonization by Group A *Streptococcus. J. Infect. Dis.* 128:527-32, 1973.
2. Fantinato V, Zelante F. *Streptococcus salivarius*: detecção de cepas produtoras de substâncias semelhantes a bacteriocina contra algumas bacterias bucais. *Rev. Microbiol.* 22(1):1-6, 1990.
3. Hardie J M, Bowden G H. Physiological classification of oral viridans streptococci. *J. Dent. Res.* 55 (special issue A):166-76, 1976.
4. Jacob F, Lwoff A, Simminovitch A, Wollman E. Definition de quelques terms relatifs a la lysogenie. *Ann. Inst. Pasteur. (Paris)* 84:222-24, 1953.
5. McCarthy C, Snyder M L, Parker R B. The indigenous oral flora of man. I. The newborn to the 1 year old infant. *Arch. Oral. Biol.* 10:61-70, 1965.
6. Pichichero M E. Explanations and therapies for penicillin failure in streptococcal pharyngitis. *Clin. Pediatr. (Phila)*. 31(11):642-49, 1992.
7. Roos K, Grahn E, Holm S E, Johansson H, Lind L. Interfering alpha-streptococci as a protection against recurrent streptococcal tonsillitis in children. *Int. J. Pediatr. Otorhinolaryngol.* 25(1-3):141-48, 1993.
8. Sahl H G. Gene-encoded antibiotics made in bacteria. Antimicrobial peptides. Wiley, Chichester (Ciba Foundation Symposium). 186:27-53, 1994.
9. Sanders C C, Sanders W E, Enicin: An Antibiotic Produced by *Streptococcus salivarius* That May Contribute to Protection Against Infections Due to Group A Streptococci. *J. Infect. Dis.* 146(5):683-90,1982.
10. Sanders, C C, Sanders W E, Harrowe D J. Bacterial Interference: Effects of Oral Antibiotics on the Normal Throat Flora and Its Ability to Interfere with Group A Streptococci. *Infect. Immun.* 13(3):808-12, 1976.
11. Sanders E. Bacterial interference. I. Its ocurrence among the respiratory tract flora and characterization of inhibition of group A streptococci by viridans streptococci. *J. Infect. Dis.* 120:698 707, 1979.
12. Tagg J R , Ragland N L. Applications of BLIS typing to studies of the survival on surfaces of salivary streptococci and staphylococci. *J. Appl. Bacteriol.* 71:339-42, 1991.