

Salivary interleukin-1 β and *Streptococcus mutans* level in oral cavity

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Abstract

Many studies have revealed correlation between serum or salivary immunoglobulin concentration and the progress of caries. Pathogens residing in the oral cavity can stimulate immune cells to produce cytokines. Antibacterial cellular reaction of T-cells in course of caries is related to the expression of superficial antigens of *Streptococcus mutans*. Interleukin-1 β (IL-1 β) – one of the most important cytokines in the human immune system – plays pivotal role in the anti-bacterial defense system. The aim of the presented study was to assess the IL-1 β level in saliva, as well as to evaluate the *Streptococcus mutans* prevalence in the oral cavity. The statistical analysis of results revealed a relevant correlation between salivary IL-1 β concentration and *Streptococcus mutans* level in oral cavity, thus suggesting the IL-1 significance in caries etiopathogenesis or denoting the modulation of cytokine concentration by bacterial antigens.

Key words: dental caries, *Streptococcus mutans*, host response, interleukin-1 β .

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Introduction

Contemporary scientific evidence unequivocally points out to the bacterial character of dental caries. Still the multidirectional assays on caries pathogenesis suggest a necessity to consider the role of immunological system in dental caries etiology. Especially interesting are the studies concerning the immunocompromised patients and invalid immune system functions [1-3]. On the other side it is common knowledge that the potentially cariogenic bacteria do not lead to carious disease in every case. The bacterial antigens can affect host organism, provoking immune response. Many pathogens residing in the oral cavity can stimulate immune cells to produce cytokines. Interleukin-1 β – one of the most important cytokines in the human immune system – plays pivotal role in the anti-bacterial defense system.

The aim of the presented study was to assess the IL-1 β level in saliva, as well as to evaluate the *Streptococcus mutans* prevalence in the oral cavity.

Material and methods

The study group comprised 86 patients aged 17-56 years, including 52 women (aged 17-56 years) and 34 men (aged

19-56 years). Interleukin-1 concentration (IL-1 β) was assessed by means of commercially available qualitative enzymatic Quantikine test (R&D System Europe Ltd., Abingdon, Oxon, United Kingdom), based on the immunoenzymatic ELISA assay, according to the manufacturer's instruction. The test specimens – mixed unstimulated saliva – were collected two hours after breakfast, then frozen and stored in -70°C . Once defrosted, each specimen was centrifuged at 1000 g for 20 min. The concentration of assessed cytokine was expressed in pg/ml. *Streptococci viridans* (mutans streptococci) were assessed in saliva collected for about 5 min upon stimulation by chewing paraffin tablet. The bacteriological assay was performed by means of commercially available medium Dentocult SM – Strip Mutans (Orion Diagnostica). The study material was collected and inoculated according to the manufacturer's recommendations. The results were classed according to the specific key, provided with the diagnostic sets mentioned above. The following classes of Dentocult SM assay were distinguished: 10^3 (class 0 – uncountable); $< 10^5$ (class 1), 10^5 - 10^6 (class 2), $> 10^6$ (class 3).

Statistical analysis: to evaluate the correlation between clinical and immunological results the Pearson's r test was used.

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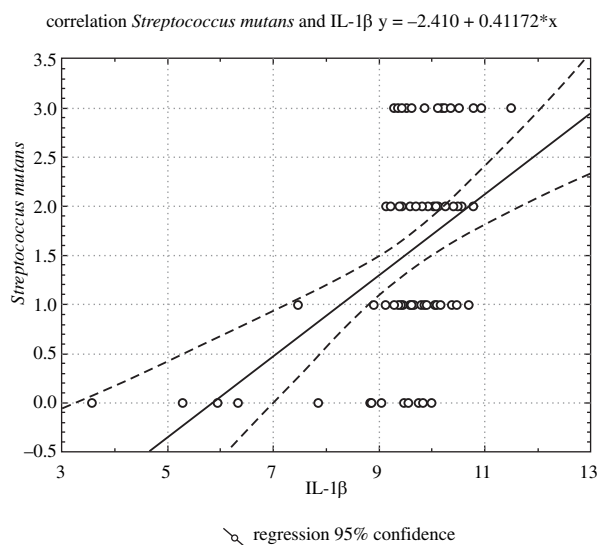


Fig. 1. Correlation between IL-1 β concentration in saliva and the level of *Streptococcus mutans* in the oral environment

Ethics

The patients and persons from control group gave a consent after details of the protocol were fully explained. The protocol of the study was conforms to the ethical guidelines of the World Medical Association Declaration of Helsinki.

Results

The presence of Streptococci viridans (mutans streptococci) was stated in 71 out of 86 analyzed salivary samples (82.56% patients). In 15 patients no countable amount of *Streptococcus mutans* was observed. Low bacteria levels (class 1) were recorded in 27 patients (31.40%), while abundant growth (class 2 and 3) was observed in 28 (32.56%) and 16 (18.60%) participants, respectively. In most of the patients ($n = 76$; 88.37%) the IL-1 β salivary concentration belonged to the range of 9 to 11 pg/ml. The IL-1 β salivary concentration in the other patients varied between 3 and 9 pg/ml ($n = 9$; 10.47%), while in only one person it exceeded 11 pg/ml (amounting to 11.50 pg/ml). The statistical analysis of study results revealed significant correlation between IL-1 β salivary concentration and *Streptococcus mutans* level in oral cavity: Pearson coefficient $r = 0.47252$; $P < 0.05$ (Fig. 1).

Discussion

Human reaction to the invasion of cariogenic bacteria is mainly based on humoral immune response [4, 5]. Many studies have revealed correlation between serum or salivary immunoglobulin concentration and the advancement of caries [6, 7]. Still the defense factors against caries probably include cellular immune response as well. Antibacteri-

al cellular reaction of T-cells in course of caries is related to the expression of superficial antigens of *Streptococcus mutans*, mainly class I/II proteins and serotype f rhamnose-glucose polysaccharide, as well as *Streptococcus mutans*-derived glucosyltransferase D [8]. Cellular immune response (T-cells), similarly to humoral immune response (B-cells), increases when the cell membrane of lymphocytes gets bound to glucosyltransferase D [9]. The studies concerning these factors aimed at developing dental caries vaccine. Yet these attempts remained unsuccessful, mainly because of numerous side effects. Eventually the adverse effects surpassed expected benefits. The trials on this area remain still in the experimental phase. In HIV-positive children a significant correlation has been observed between increased DMFT/dmft values and decreased CD4/CD8 proportion (below 0.5) [1, 3]. Children of lower caries frequency present significantly higher levels of cellular immune response indicators as compared to the children exhibiting high caries frequency [10]. The correlation between caries advancement and major histocompatibility complex (MHC) antigens has also been subject of studies [11]. The multidirectional character of discussion concerning the role of immune system in dental caries etiopathogenesis indicates necessity for further studies on this field [12, 13]. An especially important element of antibacterial defense are cytokines [14-16]. The presented study is the first to discuss the possible correlation between cariogenic bacteria prevalence in oral cavity and salivary IL-1 β concentration. It cannot be excluded that the oral cavity colonization by cariogenic bacteria is secondary to the genetically determined specific cytokines concentration. On the other hand, many antigens present in the oral cavity can stimulate cytokine production. In the presented study in most of the patients ($n = 76$; 88.37%) the IL-1 β salivary concentration varied between 9 and 11 pg/ml, that could be recognized as a physiological range of this cytokine in the studied population. The statistical analysis of results revealed a relevant correlation between salivary IL-1 β concentration and *Streptococcus mutans* level in oral cavity, thus suggesting the IL-1 significance in caries etiopathogenesis or denoting the modulation of cytokine concentration by bacterial antigens. It's hard to formulate an unambiguous conclusion. The susceptibility or immunity against caries is definitely determined by immune forces of human body. The results presented above are encouraging and raise hope for possible new perspectives in dental caries prophylaxis and treatment.

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