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Evaluation of sICAM-1 Concentration in Saliva and Blood Serum in Patients with Periodontitis

Ocena stężenia sICAM-1 w ślinie i surowicy krwi pacjentów z zapaleniami przyzębia

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Abstract

Background. Cell Adhesion Molecules (CAM) play an important role in regulation of immune response providing regular course of interaction between immunocompetent cells as well as circulation of these cells between blood and lymphatic system. They are essential for leukocytes adhesion to the cells of endothelial vessels as well as to extracellular matrix, in this way regulating migration and recirculation of immune system cells.

Objectives. Determination of the concentration of sICAM-1 in saliva and blood plasma in patients with periodontitis and in the group of healthy people and comparison of concentration of sICAM-1 in saliva and blood serum in patients suffering from chronic periodontitis and aggressive periodontitis (group I and II).

Material and Methods. Levels of sICAM-1 were measured using a sandwich enzyme linked immunosorbent assay ELISA. The values of sICAM-1 concentration were given in ng/ml of the examined material.

Results. The examinations showed statistically significant higher concentrations of sICAM1 in both saliva and blood serum of patients with periodontitis in comparison to healthy people (group III). No statistically significant differences in the concentration of sICAM-1 in saliva between group I and II were shown.

Conclusions. Elevated concentration of sICAM-1 in saliva and blood serum in patients with periodontitis is in favour of immunoinflammatory reaction in pathophysiology of periodontitis. Determination of sICAM-1 concentration in saliva and blood serum may be useful as accessory investigation in the diagnostics of periodontal diseases (*Dent. Med. Probl.* 2005, 42, 2, 223–226).

Key words: periodontitis, saliva, blood plasma, adhesion molecule sICAM-1.

Streszczenie

Wprowadzenie. Częsteczki adhezyjne (CAM) odgrywają ważną rolę w regulacji odpowiedzi odpornościowej, zapewniając prawidłowy przebieg interakcji między komórkami immunologicznie kompetentnymi i cyrkulacją tych komórek między krwią a układem limfatycznym. Są bardzo istotne dla przylegania leukocytów do komórek śródbłonna naczyń i macierzy zewnątrzkomórkowej, tym samym regulując migrację i recyrkulację komórek układu immunologicznego.

Cel pracy. Oznaczenie stężenia sICAM-1 w ślinie i w surowicy krwi pacjentów z zapaleniami przyzębia i w grupie osób zdrowych oraz porównanie stężenia sICAM-1 w ślinie i w surowicy krwi pacjentów cierpiących na przewlekłe i agresywne zapalenia przyzębia (grupa I i II).

Materiał i metody. Częsteczkę adhezyjną sICAM-1 oznaczano ilościowo metodą immunoenzymatyczną ELISA. Stężenie badanej częsteczki podawano w ng/ml badanego materiału.

Wyniki. Stwierdzono statystycznie istotne wyższe stężenia sICAM-1 zarówno w ślinie, jak i w surowicy krwi u pacjentów z zapaleniami przyzębia w porównaniu z osobami zdrowymi (grupa III). Nie wykazano statystycznie istotnych różnic w zawartości sICAM-1 w ślinie między grupą I i II.

Wnioski. Podwyższone stężenie sICAM-1 w ślinie i w surowicy krwi u pacjentów z zapaleniami przyzębia przemawia za rolą odczynu immunozapalnego w patofizjologii periodontopatii. Oznaczanie stężenia sICAM-1 w ślinie i w surowicy krwi może być przydatne jako badanie dodatkowe w diagnostyce chorób przyzębia (*Dent. Med. Probl.* 2005, 42, 2, 223–226).

Słowa kluczowe: zapalenie przyzębia, ślina, surowica krwi, częsteczka adhezyjna sICAM-1.

Polymorphism of the inflammation of the paradontium tissues shows that the pathomechanism of the processes is complex. It may be assumed that immunological mechanism plays an important role here. The main objective of the study is to determine the activity of the cells of the immune system in the diseases of paradontium. The development of methods of analysis of immunological mechanisms, interaction between immunocompetent cells and tissue cells of the paradontium as well as binding of adhesive molecule expression with the presence of suitable bacteria and inflammatory activity of cytokines in the disorders of paradontium may provide one with essential information for better understanding of etiopathogenesis of periodontopathy [1, 2].

Immunoregulation of inflammatory reactions is related to the function of cells regulating these reactions that is T lymphocytes. Cell Adhesion Molecules (CAM) play an important part in this process, especially in its initial stage.

These molecules take part not only in migration of immunocompetent cells to tissues – the place where reaction takes place – but also in their maturation, differentiation and activation. They play an important role in regulation of immune response providing regular course of interaction between immunocompetent cells as well as circulation of these cells between blood and lymphatic system. They are essential for leukocytes adhesion to the cells of endothelial vessels as well as to extracellular matrix, in this way regulating migration and recirculation of immune system cells [1, 3].

These are glycoproteins found on the surface of cell membranes acting as receptors enabling mutual adhesion of cell to cell. Due to their chemical structure, adhesive molecules were divided into 3 groups: selectines, integrines and immunoglobulinosimilar ones [1, 3, 4].

Adhesive molecules regulate immune response of the organism that is why both disorders of the expression as well as their deficiency may be the cause of severe disturbances in the functioning of the immune system.

Regardless the well known cellular forms of adhesive molecules, their soluble forms were shown. It relates mainly to molecules belonging to two groups: selectines and immunoglobulinosimilar molecules.

Seth et al. [5] showed the presence of soluble forms of sICAM-1 using immunoblotting method in 1991. This was confirmed by Rothlein et al. [6] using ELISA method.

The function of soluble adhesive molecule ICAM-1 has not been recognized so far. What is known, however, is the fact that sICAM-1 molecule is able to bind with LFA-1 receptor. On the

basis of this, it is believed that by blocking this receptor, this molecule may possess anti-inflammatory properties that is suppress adhesion and/or cause deadhesion of leukocytes.

Regardless the unclear role of sICAM-1, it is known that the presence of this molecule in elevated concentrations in blood serum or other body fluids is related to active inflammatory process of varying base [3, 5, 7].

Assuming that immunoinflammatory reaction plays a role in pathophysiology of the diseases of paradontium, it is advisable to analyze the concentrations of sICAM-1 in both saliva and blood serum of people with periodontitis.

Material and Methods

55 healthy people being under no antibiotic treatment six months before the examination were qualified for laboratory tests. Periodontitis was diagnosed in 35 patients in two forms: chronic periodontitis and aggressive periodontitis.

The group included 35 women and 20 men at the age between 28–65 years (the average age 43.3). The subjects were divided into 3 groups: group I – 20 people at the age between 28–65 (the average age 50.7 years) including 11 women and 9 men with chronic periodontitis; group II – 15 people at the age between 28–45 (the average age 37.3 years) including 9 women and 6 men with aggressive periodontitis; group III – 20 people at the age between 29–50 (the average age 37.6 years) including 10 women and 10 men without any diseases of paradontium – control group.

Saliva taken by the use of the method of absorption using Sarsted 'Salivette' set and blood serum were used as study material. The blood was taken from the ulnar vein (2 ml) using disposable instruments.

The material underwent centrifugation for 10 min with 3000 g and as then frozen at a temperature -70°C and kept until the necessary amount of samples were collected. After being defrosted, the plasma was 20 times diluted while saliva was examined without dilution.

Adhesive molecule sICAM-1 was determined quantitatively with the use of immunoenzymatic ELISA method on R & D SYSTEMS, QUANTIKINE, micro pads according to the manufacturer's instructions. The values of sICAM-1 concentration were given in ng/ml of the examined material.

The results were statistically worked out. A non-parametric U Mann-Whitney test was used to compare the groups since the examined parameters slightly differed from the original disintegration.

Significance level $p(\alpha) < 0.05$ was used as the one applied in medical studies.

Results

In all examined patients, determined titers of sICAM-1 were detected both in saliva and blood serum.

The average values of sICAM-1 concentrations in saliva and blood serum were the highest in group I in patients with chronic periodontitis and were 4.8 ng/ml in saliva and 427.3 ng/ml in blood plasma. In group II on the other hand the average values of sICAM were 2.16 ng/ml in saliva and 296.4 ng/ml in blood serum that is half times lower than in group I, and higher than group III (control group). In group III the average concentrations of examined molecule were 1.39 ng/ml in saliva and 213.8 ng/ml in blood serum.

The examinations showed statistically significant higher concentrations of sICAM1 in both saliva and blood serum of patients with periodontitis in comparison to healthy people (Fig. 1 and 2). No statistically significant differences in the concentration of sICAM-1 in saliva between group I and II were shown (Fig. 1).

Discussion

The possibility of monitoring concentrations of circulating isoforms of adhesive molecules in body fluids increased along with the accessibility to modern immunological methods including immunoenzymatic ELISA test [3, 7–9].

It must be admitted that clinical diagnostics was enriched with a new parameter. Its value seems to be significantly important in the diseases of immune pathogenesis [3, 5, 8].

Cuida et al. [8] determined concentration of sICAM-1 both in blood serum and saliva of the patients suffering from Sjögren syndrome, the disease in which immunological mechanisms play an important role. The authors used ELISA test to determine concentrations of examined molecule in saliva. The average values of sICAM-1 in ill patients were 28.3 ± 20.8 ng/ml. Determined titers of sICAM-1 were not found in saliva of healthy people (control group), (sensitivity of the method was 14 ng/ml and the values below this level were not determined).

In blood serum on the other hand, the average values of sICAM were 427.1 ± 253.3 ng/ml in patients with Sjögren syndrome and 195.6 ± 30.7 ng/ml in control group (healthy people) [8]. The findings in both control groups are compar-

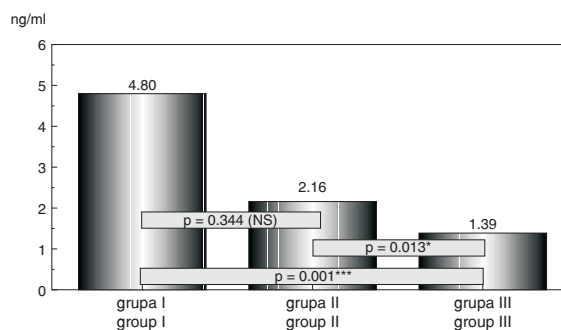


Fig. 1. Mean levels of sICAM-1 in saliva in all groups

Ryc. 1. Średnie wartości stężenia sICAM-1 w ślinie w poszczególnych grupach

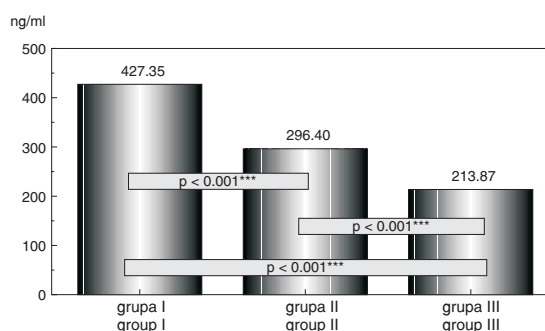


Fig. 2. Mean levels of sICAM-1 in blood serum in all groups

Ryc. 2. Średnie wartości stężenia sICAM-1 w surowicy krwi w poszczególnych grupach

ble. The value of sICAM-1 concentration in blood serum in group of people with healthy paradontium (group III) was comparable to the average value of sICAM-1 for healthy people presented in the literature [6–9].

Koundouros et al. [9] compared concentrations of sICAM-1 in blood serum of patients with paradontitis and in those without periodontitis. Study group was also divided into smoking and non-smoking one. The highest concentrations of sICAM-1 in blood serum were noticed in smokers while the lowest in healthy non-smokers.

The only comparable results of sICAM-1 in blood serum of healthy non-smokers were those presented by Koundouros et al. [9] in his own study 213.8 ng/ml and his study 210.7 ng/ml.

Evaluation of sICAM-1 content in blood serum was also carried out by Rothlein et al. [6]. The authors determined concentrations of adhesive molecule sICAM-1 in blood serum of healthy people as well as those suffering from leukocytes adhesion deficiency (LAD). Patients suffering from this disorder migration of neurophilic granulocytes through the wall of the vessel is disturbed. As a result, granulocytosis with the lack of these cells in inflammatory focus is present in peripheral blood.

The average values of sICAM-1 for healthy people are present in 100–200 ng/ml and in patients suffering from LAD from 200–700 ng/ml. Similar values of sICAM-1 concentrations in blood serum were obtained in control group of own study (healthy people) and were 213.8 ng/ml.

Mole et al. [7] determined concentrations of sICAM-1 in gingival fluid in patients suffering from gingivitis (G), adult periodontitis (AP), rapidly progressive periodontitis (RPP) and in the group of healthy people.

The values of sICAM-1 concentrations in gingival fluid were higher in people with abundant plaque and severe inflammation and were 240 ± 49 ng/ml in comparison to the patients without dental plaque, when they were 113 ± 37 ng/ml.

Mole et al. [7] obtained the highest values of sICAM-1 in gingival fluid in patients with adult periodontitis (AP), lower in the group with rapidly progressive periodontitis (RPP), and the lowest in the group of healthy people that is 110 ± 41 ng/ml so similarly to own studies.

Elevated concentration of sICAM-1 in saliva and blood serum in patients with periodontal diseases is in favour of immunoinflammatory reaction in pathophysiology of periodontitis. Determination of sICAM-1 concentration in saliva and blood serum may be useful as accessory investigation in the diagnostics of periodontal diseases.

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