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## Y. Kuzenko, A. Romanyuk, \*Y. Lindina, O. Hudymenko T-LYMPHOCYTES AND PERIODONTAL INFLAMMATION (SHORT REPORT)

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**Kuzenko Y., Romanyuk A., Lindina Y., Hudymenko O.** T-lymphocytes and periodontal inflammation (short report) // Український морфологічний альманах. – 2014. – Том 12, № 4. – С. 33-35.

Object. The object of this study was to analyze the expression of CD3 in periodontal inflammation

**Methods.** 27 patients (giant-cell epulis) and 30 patients (acute and chronic inflammations) evaluated for expression of CD3 by immunohistochemistry.

**Results.** CD3 no expressed in giant-cell. By immunohistochemistry, 0% of giant-cells were positive for CD3, whereas only 23.28% of infiltrative cells were positive for CD3 (P<0.05). As far as immunoexpression of CD3 in acute inflammation, 32,7±2,43% was positive cells. Cells infiltration, immunoreactivity of CD3 was 56.2±8,6% (P<0.05) in chronic process.

**Conclusion.** Chronic periodontopathies have been characterized by the presence in the periodontal connective tissue of an abundant chronic T cells inflammatory infiltrate.

**Key words:** CD3, immunohistochemistry, oral giant-cell epulis, inflammation.

**Кузенко Е., Романюк А., Лындина Ю., Гудыменко Е.** Т-лимфоциты и воспаление пародонта (краткие сведения) // Український морфологічний альманах. — 2014. — Том 12,  $\mathbb{N}$  4. — С. 33-35.

Объектом данного исследования является анализ экспрессии CD3 в пародонте при воспалении.

**Методы.** 27 пациентов с гигантоклеточным эпулисом и 30 пациентов с острым и хроническим воспалением пародонта оценивали на наличие CD3 положительных клеток иммуногистохимии.

**Результаты.** Гигантские клетки не были CD3 положительными, тогда как только 23,28% клеток инфильтрата в гигантоклеточном эпулисе были CD3 положительными (P < 0,05). При остром воспалении 32,7  $\pm$  2,43% клеток клеточного инфильтрата были CD3 позитивными. CD3 + иммунореактивность была 56,2  $\pm$  8,6% в клеточном инфильтрате (p < 0,05) при хроническом процессе.

**Заключение.** Хроническое воспаление пародонта характеризуется наличием в соединительной ткани обильной Т-клеточной воспалительной инфильтрации.

**Ключевые слова:** CD3, иммуногистохимия, эпулис, воспаление.

**Кузенко Є., Романюк А., Линдіна Ю., Гудименко О.** Т-лімфоцити і запалення пародонту (короткі відомості) // Український морфологічний альманах. — 2014. — Том 12, N 4. — С. 33-35.

Об'єктом даного дослідження є аналіз експресії СD3 позитивних клітин в пародонті при запаленні.

**Методи.** 27 пацієнтів з гігантоклітинним епулісом і 30 пацієнтів з гострим і хронічним запаленням оцінювали на наявність CD3 позитивних клітин імуногістохімічно.

**Результати.** Гігантські клітини не є CD3 позитивними, тоді як тільки 23,28% клітин інфільтрату в гігантоклітинному епулісі були CD3 позитивними (P < 0,05). При гострому запаленні клітинний інфільтрат мав 32,7  $\pm$  2,43% CD3 позитивних клітин. CD3 иммунореактивность була 56,2  $\pm$  8,6% у клітинному інфільтраті (p < 0,05) при хронічному запаленні.

**Висновок.** Хронічні запалення пародонту характеризується наявністю в сполучній тканині рясної Т-клітинної запальної інфільтрації.

Ключові слова: СD3, імуногістохімія, епуліс, запалення.

**Introduction.** The most common and most investigated dental diseases is periodontal inflammation. The major cause of gingivitis is an accumulation of microbial plaque in and around the dentogingival complex, which, when removed, results in complete resolution of the inflammatory lesion [1].

Page et al revealed the early stages of gingival inflammation, pro-inflammatory cytokines secreted by activate T-lymphocytes, macrophages and other cells (e.g. fibroblasts, epithelial and endothelial cells) [2].

T-lymphocytes is specifically related to CD3 protein complex within the cellular membrane of T-lymphocytes. CD3 antigen is presented in the membrane of all mature T-cells and natural killer lymphocytes (NK) being absent in other types of lymphocytes [3]. CD-3+ inflammatory cells gingival was correlated with an increase of collagen loss significantly [4].

The aim of this study is to determine the presence of T-lymphocytes cells in periodontal inflammation, and giant-cell epulis

**Methods.** The study samples consisted of the periodontal and epulis tissues of patients. The subjects were divided into two equal groups:

Patient Group (Group 1) consisted of 27 people who had a morphologically diagnosis of giant cell granuloma.

Control group (Group 2) consisted of 30 patients who had died in Sumy Regional Hospital. The patients had various diagnoses (not atherosclerotic ones).

The study population included 27 patients with giant-cell epulis. Only patients with available tissue represent a subset of the overall study cohorts.

Immunostainings for CD3 was performed on formalin-fixed (pH 7,4), paraffin-embedded thyroid tissue sections using mouse monoclonal anti- CD3 (Thermo Fisher Scientific UK). Briefly, 4μm thick tissue sections were dewaxed in xylene and were brought to water through graded alcohols. Antigen retrieval was performed by microwaving slides in 10mM citrate buffer (pH 6.2) for 30 min at high power, according to the manufacturer's instructions. To remove the endogenous peroxidase activity, sections were then treated with freshly prepared 1.0% hydrogen peroxide in the dark for 30 min at 37° C temperature. Nonspecific antibody binding was blocked using blocking serum. The sections was incubated for 30 min, at 37° C temperature, with the primaries antibodies against CD3 diluted 1:100 in phosphate buffered saline (PBS) pH 7.2, After washing 3 times with PBS. Anti-(mouse IgG)horseradish peroxidase conjugate (1:40 000 dilution) was used for the detection of the CD3 primaries antibodies, sections were then incubated for 20 min, at 37° C temperature. The colour was developed by DAB.

Appearance of positive factors was detected semiquantitatively by counting of positive giant cells in visual field.

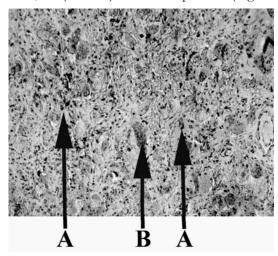
The data was analysed using STATISTICA 8.0 software, user version STA862D175437Q. The results are presented as average values (±SD). The K-S test was used in order to evaluate the normality of the data. Also, the Student method was used to perform simple comparative analysis. A value ofp < 0.05 was considered significant.

**Results.** Groups 1 and 2 men and women were prevalence in the 30- to 70-year-old-age. Group 1 found the lower jaw (55%) than in the

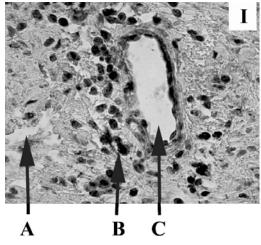
upper jaw. In the group 2 patients divided 13 with acute and 17 with chronic inflammations. Staining by G-E we studied previously.

CD3 expressed in giant-cell epulis. Expression of CD3 in giant-cell epulis shown in Fig. 1. By immunohistochemistry, 0% of giant-cells have been positive for CD3, whereas only 23.28% of infiltrative cells were positive for CD3 (*P*<0.05).

The immunoexpression of CD3 (Group 2) has confirmed by the presence of brown stained cytoplasm in cells infiltration. In general, CD3 staining was more intense in the T-cells. As far as immunoexpression of CD3 in acute inflammation (Fig-I. 2), 32,7±2,43% was positive cells. Cells infiltration, immunoreactivity of CD3 was 56.2±8,6% (P<0.05) in chronic process (Fig 2-II).



**Fig. 1.** Expression of CD3 proteins in giant-cell epulis (x100 magnification) A – T-cells CD3 "+", B – giant cells CD3 "-"



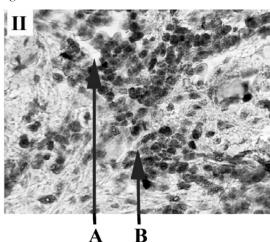


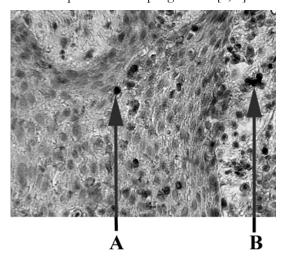
Fig. 2. Expression of CD 3 in gingival, group 2, (x400 magnification) I acute inflammation A - pericellular and perivascular edema, B-T "+" blood cells infiltration, C - blood vessel. II chronic process A - zone of edema B - T cells infiltration.

The results of the chronic process shown in Fig 3. Widespread T-cells reactivity in epithelium (Fig. 3). T- lymphocyte reactivity in epithelium was evidence cells migration into oral cavity.

**Dictation.** The inflammatory infiltration of

periodontitis is composed of mononuclear cells, mainly mononuclear phagocytes and lymphocytes. T lymphocytes predominate in the nonprogressive stable periodontal lesion in which CD4-H cells predominate over the CD8-type. The

proportion of B-lymphocytes and plasma cells increases as periodontitis progresses. [5, 6]



**Fig. 3.** Expression of CD3 in gingiva, group 2, chronic process (x400 magnification) A - T-cells in epithelium, B T-cells infiltration.

T cells carry out several functions within the immune response: lyses the cells that express noself molecules on their surface, regulates the immune response, mediates the reactions of delayed hypersensitivity, synthesizes the most part of lymphokines, stimulates the differentiation of Blymphocytes towards plasma cells, etc

Douglas D. et all studied modulation of the JAK3 pathway is critical for specific type 1 cytokine receptors, which possess common chains and can modulate CD3 mediated activation, leading to T cell anergy expression is a key signaling intermediate for receptor signaling in both T and NK cells, and the JAK/STAT pathway is also critical for T regulatory cells and NK cell activation and function. [7]

**Conclusion.** Chronic periodontopathies have been characterized by the presence in the periodontal connective tissue of an abundant chronic T cells inflammatory infiltrate.

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