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FEATURES OF STRUCTURAL ALTERATION IN PERIODONTAL COMPLEX COMPONENTS AT THE EARLY STAGE OF THE EXPERIMENTAL BACTERIAL-IMMUNE PERIODONITIS DEVELOPMENT A.YE. DEMKOVYCH I. Horbachevsky Ternopil State Medical University, Ukraine, Ternopil

ТАЖРИБАДА БАКТЕРИАЛ-ИММУН ПАРОДОНТИТ РИВОЖЛАНИШИНИНГ ЭРТА БОСҚИЧИДА ПАРОДОНТАЛ КОМПЛЕКС СТРУКТУРАЛИ АЛМАШИНУВ КОМПОНЕНТЛАРИНИНГ ХУСУСИЯТЛАРИ

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ОСОБЕННОСТИ СТРУКТУРНОЙ ПЕРЕСТРОЙКИ КОМПОНЕНТОВ ПАРОДОНТАЛЬНОГО КОМПЛЕКСА НА РАННЕМ ЭТАПЕ РАЗВИТИЯ ЭКСПЕРИМЕНТАЛЬНОГО БАКТЕРИАЛЬНО-ИММУННОГО ПАРОДОНТИТА

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Пародонтдаги яллигланиш жараёнлари стоматологик касалликлар ичида кенг тарқалган булиб, кайталаниб кечиш билан характерланади ва беморлар хаёт сифатини нихоятда пасайтиради. Пародонтал комплекс тўқимасида яллигланиш жараёни ривожланишида мавжуд бўлган структурали ва патоморфологик ўзгаришларнинг нисбийлиги ушбу касалликда деструктив белгиларнинг ривожланиш босқичларини тўлиқ ифодаламайди, шунинг учун бу касаллик чуқур тадқиқотга мухтож. Тадкикот максади – тажрибада бактериал-иммун пародонтит ривожланишининг 7-чи суткасида пародонтал комплекс структурали компонентларининг хусусиятларини ўрганиш. Тадқиқот натижасида соглом хайвонларда хусусий пластинканинг бириктирувчи тукимаси коллаген толалардан иборат эканлиги, баланд бириктирувчи тўкимали сўргичлар хосил килиши ва эпителийга зич ёпишиши аниқланди. Гистологик тахлил натижасига кўра, тажрибадаги бактериал-иммун пародонтит ривожланишининг 7-чи суткасида бутун пародонт бүйлаб типик яллигланиш жараёни тарқалганлиги маълум бўлди. Аммо бу жараённинг яққоллиги пародонтнинг структурали компонентларида хар хил эканлиги аниқланди. Хўжайра элементлари ичида кўпинча фибробластлар учради, кам холатда гистиоцитлар, лимфоцитлар, якка плазмоцитлар ва моноцитлар учради. Тадкикотнинг бу боскичида фибробластлар миқдорининг ошиши кузатилди, булар коллаген толалар резорбция функциясини бажариб, фаол остеобластлар миқдорини камайтирди. Бу барча патоморфологик ўзгаришлар яллигланиш жараёни ривожланиши белгиларидан дарак беради.

Калит сўзлар: Пародонтит, яллигланиш, периодонт, пролиферация, коллаген толалар.

The inflammatory process in the periodontium is the most widespread among dental diseases, that is characterized by a recurrent course and significantly reduces the quality of life patients. Existing data about structural and pathomorphological changes in the tissues of the periodontal complex for the of inflammation development does not explain largely the patterns of the destructive phenomena development in this pathology. and therefore require additional research. The purpose of this study was to determine the peculiarities of structural components of periodontal complex and their changes on the 7^{th} day of the experimental bacterial-immune periodontitis development. In the study of intact animals, it was found that connective tissue of its own plate is represented by collagen fibers, forms high connective tissue papillae and closely adheres to the epithe-lium. The histological study showed that on the 7th day of the experimental bacterial-immune periodontitis development of typical and spread inflammatory reaction onto all periodontium was observed. However, the severity of this reaction was different in the structural components of the periodontium. Fibroblasts were the most often found among cellular elements, less frequently – histiocytes, lymphocytes, single plasmacytes and monocytes. It was an increase of the fibroblasts number, that perform the resorptive function of collagen fibers and reduce number of active osteoblasts. All these pathomorphological changes testified about of the inflammatory process development.

Key words: Periodontitis, inflammation, periodontium, proliferation, collagen fibers.

Introduction. The periodontal diseases by the character of clinical course concern predominantly to the chronic and complete inflammatory-destructive changes in tissues that hold teeth in the alveolar bone,

and lead to progressive growth of connective tissue. The data of epidemiological studies indicate that the currency of periodontal diseases in the world ranges from 5-20% and with age increases to 75% [1, 2]. In

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recent years, generalized inflammatory diseases of periodontium attract increased interest of researchers and clinicians, because after 35 years age they lead to extraction of teeth, increase the risk of associated systemic pathology development. It is known that exactly inflammatory processes that develop in the periodontal complex are most often the main cause of teeth loss [3, 4].

Features of the structural organization of tissues that are part of the periodontium, promote to damage with mechanical, chemical, bacterial and immune factors, and the integrality of the structure depends on maintaining appropriate level of metabolic, microcirculatory processes, neuroendocrine regulation [5, 6].

In the diagnostics of periodontal diseases to clinical and morphological research is put the task of determination degree, activity and spread of destructive process. On its base is defined an objective assessment of the severity of the injury and an adequate treatment tactic is worked out [7, 8]. In this regard, expediently an experimental study of structural changes in the soft and bone tissues of periodontium, that forms under influence of pathogenetic processes, which underlie of inflammatory and dystrophic disorders in the periodontal complex [9, 10].

At present, scientific researches have received enough material concerning structural changes as a result inflammation in periodontal tissues, but the consequentness and lawfullness of its formation are far from being studied.

The purpose of this research is to determine peculiarities of changes in the structural components of the periodontal complex in the early period (on the 7th day) of the experimental bacterial-immune periodontitis development and to give them a prognostic assessment.

Materials and methods. The experimental bacterial-immune periodontitis was induced in the experimental animals by introducing complex mixtures of microorganisms diluted in egg protein into periodontal tissue [11]. Simultaneously with the injections of the microbial pathogen, a complete Freund's adjuvant was injected in the rat's paw to enhance the immune response. The experimental studies were performed with the use of clinically healthy rats weighing 150-200 g in the conditions of vivarium. The animals were in a standard diet balanced by the main elements of nutrition. Systematically healthy rats of the same age were used as controls. The reseach was performed in accordance with sanitary-hygienic norms and GLP requirements. The experiments were conducted in conformity with the general rules and provisions of the "European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes" (Strasbourg, 1986), and the "General Ethical Principles of Animal Experimentation" (Kyiv, 2001).

For evaluation of the degree structural changes in the tissues of the maxillofacial area was used a morphological study. The experimental animals were sacrificed on the 7th day through decapitation under thiopental anesthesia. The fragments of the mandible tissues, in particular, the periodontal complex were removed, washed in saline from the blood and fixed in 10% neutral formalin solution. The material was poured in paraffin blocks. The transverse sections on a microtome were made in the thickness of 5-6 microns. The resulting preparations were stained with hematoxylin and eosin [12].

Results and discussion. Considering that histological norm of periodontal components is known and described in the literature [13, 15], however existing individual variability of tissues there is necessity to study them in the group of intact animals.

The mucous membrane of the rat's gums is covered with a multilayer plane epithelium with expressed signs of keratinization and parakeratosis besides corneal layer was uneven thickness and has view homogeneous eosinophilic strip. The usual phenomenon was small areas of superficial desquamation of the epithelium.

The granular and spinal layers located below had not layer-by-layer differentiation, as opposed to basal, which clearly defines the boundary of the epithelial lining and its own plate of the mucous membrane. The dystrophic changes of the epithelial cells in this group were presented formations with weak to moderately expressed. In such cases in the cytoplasm of the keratinocytes was visualized vacuoles of different sizes and different amounts.

The connective tissue of its own plate was represented by collagen fibers, which formed high connective tissue papillae and tightly adjacent to the epithelium. The papillary layer was presented with a loose connective tissue. The collagen fibers in deeper part had more compact, dense, location and transited into the periosteum of the cortical plate of the alveolar processes. Among the cellular elements, fibroblasts were found most frequently, lesser frequently – histiocytes, lymphocytes, single plasmacytes and monocytes (Figure 1).

The own plate was intensively vascularized, especially in the superficial layers. Numerous blood capillaries of the surface layer had narrowed lumen, almost did not contain form elements and therefore poorly visualized. The hemocapillar wall contain s plane endothelial cells that located on the basement membrane. The periodontium was represented by sidelong directed bundles of oxyphilic collagen fibers, between that layers of loose connective tissue with thin wall vessels were visualized. The cellular composition was diverse, in particular, it included epithelial and osteogenic basophils, undifferentiated mesenchymal cells, except of fibroblasts, which were the predominant type of cells (Figure 2).



Figure 1 – Structural organization of the rat's gum of the control group. Staining with hematoxylin and eosin. × 200



Figure 2 – Structural organization of the rat's periodontium of the control group. Staining with hematoxylin and eosin. $\times 200$

The cement of the tooth roots is constructed with collagen fibers, which contained layers of amorphous matter. Cellular elements of the cement appeared closer to the top of the root – cementoblasts and cementocytes. Vessels in this structure were not found.

The alveolar process, which surrounds the tooth root, represents a thin bone plate, to which the periodontal ligament attached. The bone plate, in one's turn, consist of plate bone tissue, it formed osteomas and was infiltrated with periodontal fibers, vessels and nerves.

Own alveolar bone of the alveolar wall passed into a supporting alveolar bone, covered with a cortical plate and formed by compactly composed bone plates. The osteocytes had an elongated shape and were placed individually in clearly-contoured gaps in the matrix.

The matrix itself was stained poorly but evenly. Orsene fibers had a multi-directional placement, due to which the nearest layers of the fabric had different optical properties. The presented vessels in cut were moderately blood-contented. The superficial layer of the periosteum was represented by collagen fibers, located parallely to the surface of the bone, with a small number of fibroblasts.

The deep – osteogenic layer was visualized unclearly and contained thin spindle-like osteogenic cells. The osteoclastes and osteoblasts appeared rarely. Sites between plate of spongy bones were filled in with red and yellow bone marrow rarely (Figure 3).

The analysis of tissue changes on the 7th day of the experimental bacterial-immune periodontitis was characterized by development of extensive inflammatory process during whole periodontal period. However, the severity of this reaction was different in the structural components of periodontum.

So, pathological changes in the gum mucus were found for that period. The epithelial lining, as a rule, became uneven thickness at that stage, which arose mainly due to desquamation of epithelial cells. The epithelium reacted with proliferation to the inflammatory process in a certain areas.

At the same time with noted above changes, the epithelial plate not infrequently became visually interrupted and preserved papillae of the surface layer its own plate reached to the surface of the epithelium. The keratinous r was also uneven thickness and interrupted, it was raised above the osteal layer.

Layer-by-layer differentiation of the epithelium was retained, but unclear especially in the surface layers. Dystrophic changes of the cell occured on the spot. Their cytoplasm became light, vacuolated, and the picnotic nucleus was displaced to periphery. Such changes were interpreted as so-called balloon dystrophy.

The cells with a hyperbasophilic festoon nucleus, surrounded by an empty ring-shaped space occurred in the suprabasal and basal layers. The cells with similar changes contrasted sharply with intact keratinocytes. The presence of these cells suggested strengthening of induced apoptosis.

Many lymphocytes were found among the keratinocytes. The clear line of the basal layer was blurred at the expense of inflammatory infiltration in the papillae of its own plate. In the infiltrate were dominated neutrophils. The pappilae of their own plate were different height and thickness, somewhere polished with polymorphocytic infiltration (Figure 4).

The bundles of periodontiumal collagen fibers somewhat were loosened and lost their clarity, their contours were uncleared and became homogeneous. The oxyphilic stainig somewhat changed onto basophilic. The amorphous component of its own plate, which became more enlightened and heterogeneous as compared with the previous observation period. Its areas expanded parallely.

Thus, on the base of that results, it may be consider that all signs of disorganization of connective tissue components occured. Both surface and deep layers were revealed lymphahistiocytic infiltrates with an admixture of neutrophils.

The capillaries and small blood vessels of the arterial type were uneven blood-filled with a predominance of hypertrophy and stasis. The veins were also mainly congested (Figure 5).

The bundles of periodontal collagen fibrils were somewhat loosened and lost their clarity. The layers of loose connective tissue that situated between them were enlarged both at the expense of the intercellular fluid, and at the expense of cellular elements that were found in that structure in the norm.

Somewhere in the infiltrate were found neutrophils. It should be noted that increase separately of number fibroblasts, performing the function of resorption of collagen fibres, promoted to remodeling of periodontium already at the initial stages of experimental study. The microcirculatory stream was unevenly blood-filled, with moderately expressed signs of hemodynamic disorders: stasis, erythrocytic sludge.

There were minimal changes in the cement of the tooth root for this experimental period that characterized the initial signs of disorganization of the connective tissue in the form of fibre structure violation and swelling of the collagen fibers.

Some disturbances in the alveolar bone structure after 7 days of the experiment were not found. The alveolar bone tissue in the experimental animals remained characteristic structure. The bodies of the osteocytes localized in the gaps, the walls of which were contoured somewhat weaker than in the control animals. The cells became more oval shape, the nuclei were hyperchromic, that caused condensation of chromatin. The osteoid between the gaps was homogeneous and somewhat enlightened.

The osein fibrils in the plates were clearly oriented. Only moderate blood filling was observed in the blood vessels. There were gigantic oxyphilic multinuclear cells – osteoclast localized near of the vessels in the superficial areas of the bone. The matrix around them looked slightly enlightened. The bone plates were thinned and found focal hollow – lacunar resorption. The cell nucleuses were also hypochromatic in the periosteal osteogenic layer. The numbers of active osteoblasts were decreased (Figure 6).



Figure 3 – Structural organization of the bone tissue rat's alveolar process of the control group. Staining with hematoxylin and eosin. × 200



Figure 4 – Histological structure of the rat's gum after 7 days of the experiment. Dense round cell infiltration of the mucous membrane own plate. Balloon dystrophy of keratinocytes. Staining with hematoxylin and eosin. × 100.



Figure 5 – Histological structure of the rat's gum own plate after 7 days of experiment. Swelling and homogenization of the collagen fibers, dense leukocytic infiltration, predominant venous hyperemia and leukostasis in the vessels. Staining with hematoxylin and eosin. × 200



Figure 6 – Histological structure of the rat's alveolar bone process after 7 days of the experiment. Cell proliferation of the mesenchyma. Initial signs of lacunar resorption of bone plates. Staining with hematoxylin and eosin. × 200

Conclusion: 1. Progressing inflammatory changes in the structural components of the animal periodontal complex arise for early period (on the 7th day of the study) of the experimental bacterial-immune periodontitis development that display by disorganization and destruction of connective tissue and walls of tooth alveolus's, structural reconstruc-

tion of epithelial gum lining and its own plate, cellular infiltration and microcirculatory disorders. 2. The character of pathomorphological changes may be a sign of deep destructive-dystrophic processes that form clinical picture of periodontitis with protracted course.

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ОСОБЕННОСТИ СТРУКТУРНОЙ ПЕРЕСТРОЙКИ КОМПОНЕНТОВ ПАРОДОНТАЛЬНОГО КОМПЛЕКСА НА РАННЕМ ЭТАПЕ РАЗВИТИЯ ЭКСПЕРИМЕНТАЛЬНОГО БАКТЕРИАЛЬНО-ИММУННОГО ПАРОДОНТИТА

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Воспалительные процессы в пародонте относятся к числу наиболее распространенных стоматологических заболеваний, характеризуется рецидивирующим течением и существенно снижает качество жизни пациентов. Существующие данные относительно структурных и патоморфологических изменений в тканях пародонтального комплекса в процессе развития воспаления не в полной мере объясняют закономерности развития деструктивных явлений при данной патологии, а потому нуждаются в дополнительных исследований. Целью данного исследования было изучить особенности структурных компонентов пародонтального комплекса и их изменений на 7-е сутки экспериментального развития бактериальноиммунного пародонтита. При исследовании интактных животных установлено, что соединительная ткань собственной пластинки представлена коллагеновыми волокнами, образует высокие соединительнотканные сосочки и плотно прилегает к эпителию. Гистологическое исследование показало, что на 7-е сутки развития экспериментального бактериально-иммунного пародонтита наблюдалось развитие типичной и распространенной на весь пародонт воспалительной реакции. Однако выраженность этой реакции была различной в структурных компонентах пародонта. Среди клеточных элементов наиболее часто встречаются фибробласты, реже - гистиоциты, лимфоциты, единичные плазмоциты и моноциты. На этом этапе исследования происходило увеличение количества фибробластов, которые выполняя функцию резорбции коллагеновых волокон и уменьшение количества активных остеобластов. Все эти патоморфологические изменения указывают на признаки развития воспалительного процесса.

Ключевые слова: Пародонтит, воспаление, периодонт, пролиферация, коллагеновые волокна.