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PATHOGENETIC ASPECTS OF DENTAL PULP PATHOLOGY

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ПАТОГЕНЕТИЧЕСКИЕ АСПЕКТЫ ПАТОЛОГИИ ПУЛЬПЫ ЗУБОВ

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The item offers a view on the effects of morphological, functional, and structural changes in overpulpal dentine and pulp while using multicomponent paste for treatment of deep caries and acute focal pulpitis. The material for the experiment included 13 teeth: 5 canines, eight premolars and molars (6 clinically outbred dogs aged 2–5 years). As found out, the use of the paste accelerated the dynamics of reparative dentinogenesis. The data from electronic microscopy scanning suggest optimization in inflammatory re-

action followed by normalization of the main structural components of the pulp and expressed activation of the dentinogenetic function.

Key words: pulpitis, caries, experiment, treatment

Представлены результаты морфологических, функциональных и структурных изменений в надпульпарном дентине и пульпе при использовании поликомпонентной пасты для лечения глубокого кариеса и острого очагового пульпита. В эксперимент включены 13 зубов: 5 клыков, 8 моляров и премоляров (6 беспородных собак в возрасте от 2 до 5 лет). Установлено, что при использовании разработанной пасты динамика репаративного дентиногенеза ускорялась. Электронно-микроскопические данные свидетельствуют об оптимизации воспалительной реакции с последующей нормализацией основных структурных компонентов пульпы и выраженной активизацией дентиногенетической функции.

Ключевые слова: пульпит, кариес, эксперимент, лечение

Understanding the mechanisms of teeth pulp repair remains an urgent issue within Regenerative Medicine [4, 7, 9, 12, 13]. Despite the excessive supply of medicinal interlinings (pulp caps) manufactured by Russian and foreign companies that are available at dental market currently and which are used in the treatment of deep caries and acute focal pulpitis, the rate of complications in the first 6 months after treatment of deep caries reaches 32%, while after the treatment of acute focal pulpitis it comes as high as 84% [6, 16]. This is due to inadequate estimation of the hard dentin structures, lack of knowledge about the repair mechanisms when choosing pharmacological agents, as well as improper technology of their application [1, 15, 20].

Treating dental pulp pathologies cannot be performed any better without the development, synthesis and manufacturing of specific medicinal interlining materials possessing pluripotent properties proven through experiments. The currently available medicinal compositions for the interlinings are predominantly unidirectional and are used either for dentin remineralization or to relieve the initial stages of inflammation, while only a small part of them can act as multifactorial agents modifying the structure of dentin as well as pulp components [2, 3, 5, 8].

Despite considerable interest in the study of changes in hard tissues and the pulp in the stages of disintegration under deep caries and acute focal pulpitis, there yet remain many issues that need clarification when it comes to the pathological mechanisms and particularly to proper selection of drugs and their efficacy in the treatment process [11, 14]. In this context, experimental morphological study of the effects wrought by new combined medicinal pastes on the overpulpal dentine structures and coronal pulp components under treatment of deep caries and acute focal pulpitis in teeth models of experimental animals, appears especially interesting and useful [8, 10, 19].

Aim of the study. Pathogenic assessment of reparative dentinogenesis in experimental pulpitis using multicomponent paste.

Material and Methods. This experimental study was carried out in accordance with Directive 2010/63/EU of the European parliament and the council of the European Union on protection of animals used for scientific purposes. The objects for the experimental studies were 6 clinically healthy outbred dogs aged 2–5 years, weighing from 4 to 8 kg, kept in the vivarium under standard conditions. The material for the experiment was 13 teeth: 5 canines, eight premolars and molars. The experiment was performed under intravenous anesthesia (xylazine, Rometar®), by type of acute experiment. The formation of deep cavities was performed according to generally accepted standards with carbide burs on the buccal surface, rotation speed – 20,000 rpm with saline cooling. This was followed with dissection of the tooth cavity and the surface of the pulp exposed through excavating under warm bath of chlorhexidine. The formed cavities were dried with sterile cotton swabs.

The treatment of simulated lesions in the deep dentin layers and the pulp surface layers was carried out using an elaborated combined medicinal paste, where the composition was as follows: lysozyme, oily solution of vitamin A, zinc oxide (30%), Dimexidum solution, dexamethasone, neomycin and collagen gel Collost [17, 18].

The samples from the experimental animals were taken in 7, 14 days, 1, 3 and 6 months. The teeth were removed with extracting forceps; sometimes bone blocks were cut out with jaw fragments under general anesthesia. The samples thus obtained were fixed in 10% neutral buffered formalin to be further subjected to slow demineralization in a 10% nitric acid solution. The dehydratation and paraffin-embedding were carried out using the closed-type histological processor Tissue-Tek VIP™ 5 Jr and the paraffin-embedding station Tissue-Tek® TEC™ 5 (Sakura, Japan). The blocks were used to prepare histological sections (5–7 μm thick) which were stained with hematoxylin and eosin, Alcian blue, by Van Gieson, Akimchenko and silvering by Mallori. The morphometric studies were performed using the Video Test Morphology 5.1 for Windows.

The scanning electron microscopy (SEM) was performed on a JEOL machine, Series JSM-6510, with a resolution in a high vacuum 3.0 nm (30 kV), 8.0 nm (3 kV), 15.0 nm (1 kV), magnification of x8 to x300,000 (at 11 kV or above), under an electric shift of the image up to ±50 μm, (WD=10 mm), saving the resulting images in the JPEG format.

The obtained experimental data were processed employing methods of variation statistics and Student's t-test using a software package of statistics Microsoft Excel.

Results and Discussion. Using the prepared medicinal composition for treating deep caries in a relatively short time led to significant changes in the main parameters for building the overpulpal dentine layers. However, attention is to be drawn to the dense consistency organization of a medicinal interlining. If viewed microscopically, the therapeutic paste does not always densely adjoin to dentine, thus leaving small cleft gaps (Fig. 1). As early as 7 days later, along the dentinal tubules there can be clearly seen immersion of the fine-grain substance to the depth of 30–40 μm. Throughout the area of the object, this substance shows varying density, which is due to two factors – the content of the dentinal tubules and redistribution of interstitial pressure in them under the impact of the medicinal interlining.

By Day 14 of the experiment the observed phenomena get more pronounced. The fine-grain substance is to be found 60–70 μm down from the dentin surface. All over the area under observation, the inputs into the dentinal tubules are obliterated. Moving closer to the pulp allows detecting narrowing in the dentinal tubules up to 2.14±0.42 – 1.86±0.40 μm.

In 1 month the zone of contact with the therapeutic paste revealed signs of more prominent pattern of peritubular and intertubular parts of the main substance of dentin, which was due to an increased mineral phase. Closer to the pulp no such changes were observed in the main substance of dentin; at the same time the walls of dentinal tubules had a large amount of fine-grain substance adsorbed.

1–3 months later most of the dentinal tubules were tightly obliterated with fine-grain and amorphous substance down to a depth of 60–80 μm. Changes in the surface of circumpulpar dentin could be revealed 1 month after the start of the experiment. Focal tight areas of main substance were formed on the surface of dentin.

By the term of 3 months a larger part of the circumpulpar dentin surface was covered with newly formed layer of the main substance containing fibrous structures, and fine-grain and amorphous substances. The layer thickness was 18.36±1.20 – 34.12±0.84 μm (Fig. 2).

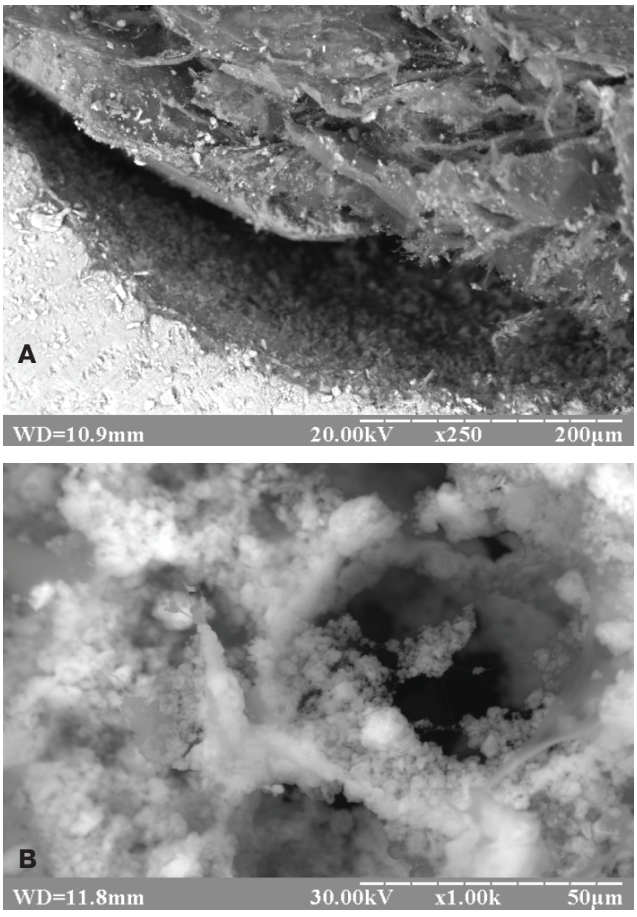

Fig. 1. Microslides. Smaller cleft gaps in between the medicinal interlining and the dentin (A). Term – 7 days. Obliteration of dentinal tubules (B). Term – 14 days. SEM

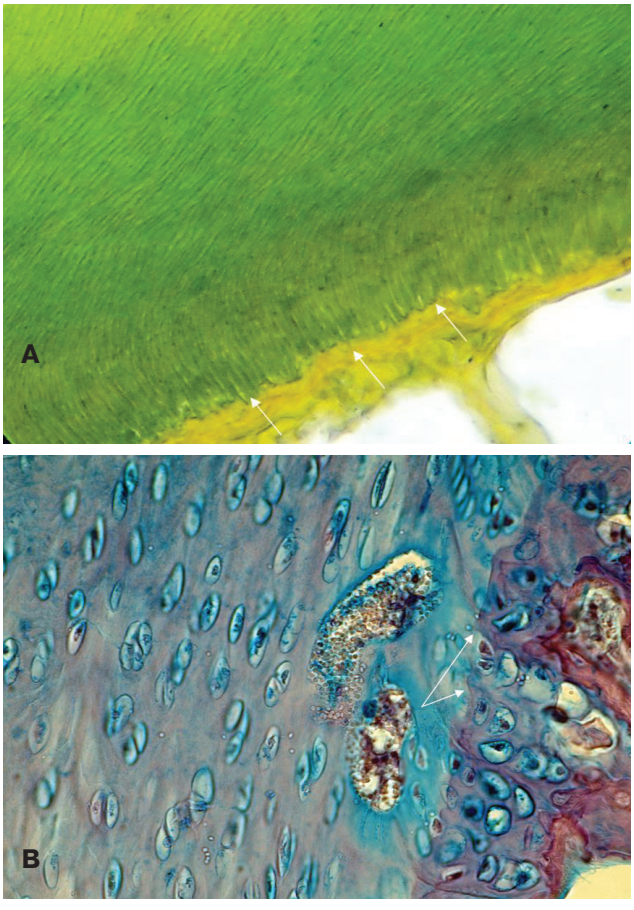

Fig. 2. Microslides. Term – 3 months. Borderline of predentin with processes of odontoblasts (A). Staining by Akimchenko. Magn. x300. Right – developing layer of replacement dentin with uneven distribution of inputs into the dentinal tubules (B). Staining with Alcian blue. Magn. x300

As for its architectonic construction, the formed layer of replacement dentin was of quality much below than that of the normal structure. First of all, the difference is manifested through uneven distribution inputs into the dentinal tubules as well as fibrous structures that lack systemic orientation.

After 6 months the replacing dentin layer significantly expands and thickens up to 28.14±0.86 – 44.42±1.04 μm. The density of fibrous structures gets increased. In some areas of the main substance fine-grain substance can be spotted. The shape and dimensions of the dentinal tubules get closer to normal ones yet the density of their arrangement over the surface is below that of the normal status (Fig. 3).

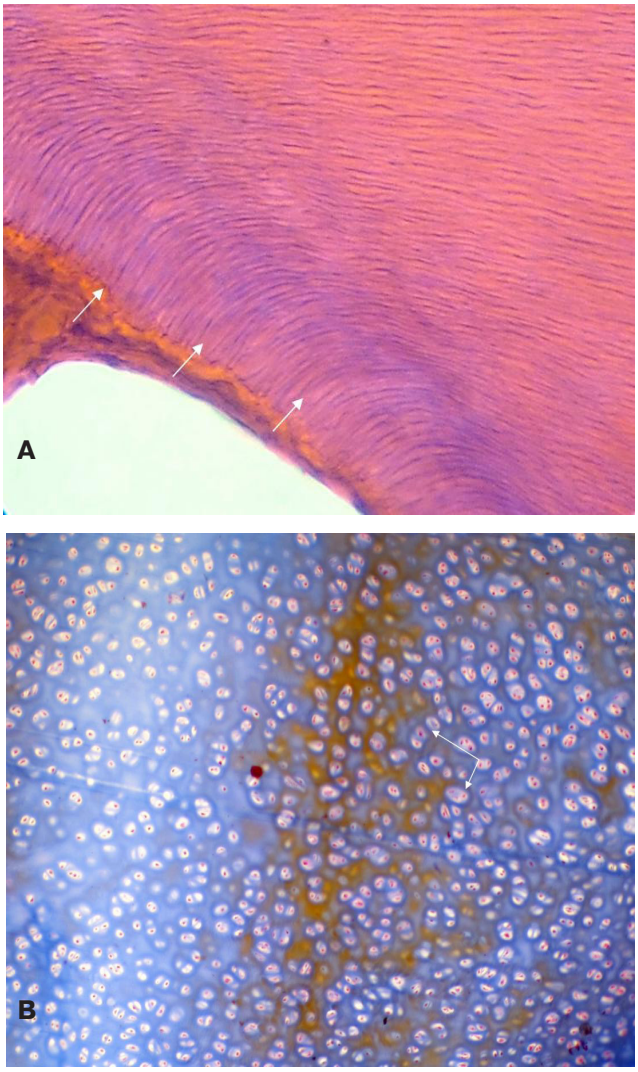

Fig. 3. Microslides. Term – 6 months. Borderline of predentin with processes of odontoblasts (A). Hematoxylin and eosin staining. Magn. x600. Peritubular dentin with processes of odontoblasts in the dentinal tubules (B). Staining with Alcian blue. Magn. x300

In the treatment of acute focal pulpitis in the shorter terms into the experiment the reaction revealed by the pulp could be described with features typical of thermomechanical factors for dissection of overpulpal dentine layers.

By Day 14 the enhancement of compensatory phenomena could be observed, unlike other series of experiment. First of all, there was clear normalization in the pulp capillaries structure, especially in the central and intermediate layers. The swelling of basic substance in subodontoblastic layer was insignificant. The number of leukocytes was reduced down to 8.04±0.82 – 11.42±0.86 in 10 out of 12 fields of view (10x12). At the same time an increase in the number of undifferentiated cells was found, which were tending to form a layer of up to 36.74±1.86 – 44.56±1.84 (8x12). The number of fibroblasts in the form of bands adjacent to the layer of undifferentiated cells was increased. The ratio of the fibrous structures against the cellular elements and the main substance was 32.12±0.44 – 36.82±0.76%, which is significantly better than the previous series of experiments using other compositions [19].

By 1 month into the experiment the layer of odontoblasts showed normalization of the structure. There were no morphological defects, micropores or cleft formations along the formed dentin layer found.

In 3–6 months no abnormalities were found in the structure of the peripheral and central pulp layers.

Given the facts above we can say that the changes in the pulp structures and in its nerve elements were of functional nature which itself manifested in activation of reactive and recovery processes while preserving the viability of the pulp. The histology of these changes showed optimization of the inflammatory process with increased levels of metabolic reactions and expressed activation of pulp cellular elements and normalization of metabolism.

The results yielded experimentally serve evidence to optimized dentinogenesis when using polycomponent medicinal composition with restoration of the overpulpal dentine and coronal pulp.

Conclusions. Our study showed that the medicinal paste designed for treating deep caries and direct pulp capping in the therapy of acute focal pulpitis has specific and, at the same time, pluripotent properties in terms of preventing inflammatory complications and enhancing the protective capacity of the pulp. Morphological evaluation of the medicinal composition application showed rapid recovery in the structure of the overpulpal dentine layers. Within 7–14 days the results attained included relief of the inflammatory response, normalization in the main components of the pulp, and significant enhancement of its dentinogenic function.

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