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EXPERIMENTAL STUDY OF THE POSSIBILITY OF USING FOLATE-POLYMER COMPOSITES FOR THE SURGICAL TREATMENT OF PERIRADICULAR BONE DEFECTS

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Abstract:

The issue of the influence of various materials for bone regeneration and its optimization remains relevant. Morpho-histological studies of the biocompatibility and bioactivity were performed on 36 white laboratory rats and have shown that folic acid in the composite material inhibited the exudative phase of inflammation. Physicochemical and biological properties of the folate-polymer composites provide an opportunity for eliminating of the periradicular bone defects and prevention of the teeth displacement.

Key words: periradicular bone defects, folate-polymer composites, implantation, biodegradation, bone formation.

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INTRODUCTION

At the present stage of the reconstructive and reparative maxillofacial surgery development, the issue of eliminating periradicular bone defects and the study of the influence of various materials for the tissue engineering, the processes of bone regeneration and its optimization remains relevant [1, 2].

Retrospective data analysis suggests quantitative advantage in the diagnosis of radicular and residual cysts (56.9% -78%) among cystic jaw formations, 63% of which were found in the maxilla, 46% – at the stage of suppuration [3, 4, 5].

From a biological point of view, the alveolar bone restoration is prevented by the following interconnected factors: retention of pathogenic microflora, inhibitory effect on the bone regeneration by the epithelium, low reparative potential of hard tissues of the periodontal complex due to the absence of the bone marrow in this area, that contains osteogenic precursor cells [6].

Debatable issues of unpredictable impact of bone replacing materials and / or membranes on the conditions and course of reparative osteogenesis remain under discussion; biological irrelevance of the timing of such materials' usage and phases of reparative osteogenesis; limited stability of synthetic calcium apatite at biomechanical loading; repeated surgical intervention to remove materials that are not subject to biodegradation [7, 8].

Alternative materials that have an effect on reparative tissue regeneration are biodegradable polymeric compositions with prolonged release of drugs [9]. The most intense processes of biodestruction, with subsequent substitution by a tissue regenerate, are prone to porous polyurethanes, which have developed surface and a large area of contact with surrounding tissues [10].

The high efficacy of surgical treatment of patients with congenital and acquired bone pathology of the maxillofacial area using biodegradable epoxy polyurethane composition fixators with sufficient and long-lasting strength characteristics has already been proven [11].

Purpose. The aim of this study was to substantiate the possibility of using folate-polymer composites for the surgical treatment of periradicular bone defects.

Materials and methods. On the basis of the Institute of Macromolecular Chemistry of National Academy of Sciences of Ukraine, composite material based on mesh polyurethane – Medical glue (TU U 21.2-05417041-024: 2013) was obtained. It has the

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ability to polymerize in a defective cavity, taking its shape and forming adhesive bond on the border of the polymer - bone - soft tissue.

Source materials of the "Medical glue": 1. Adhesive base (AB) – oligourethane diisocyanate – 2 g; 2. Polymerization accelerator (PA) - 2,4,6-tris (dimethylaminomethyl) phenol - 0.03 g; 3. Filler (F) - folic acid - 0,010 g.

The ratio of adhesive components is selected taking into account the biological effect of the filling substance (folic acid) and the quality of the polymerization of the other two components.

After polymerization, the microporous structure of the composition (Fig. 1) contributes to stability and gradual penetration of the newly formed tissues deeper as the material is biodegrading, the diffusion of biological fluids, which greatly improves the conditions for tissue regeneration [12].

Fig. 1. Macrograph of microporous structure of the folate-polymer composite sample

An overview of the scientific data on the internal structure of polyurethane composite [13] and ourselves investigations (M.A. Skoryk, V.S. Shvydchenko, 2017) obtained with the help of scanning electron microscopy (SEM) confirms the presence of pores (231 – 806 μм) and interconnected interactions (45 – 270 μм)- the "windows" between them, which likens it to natural porosity of trabecular bone (Fig. 2).

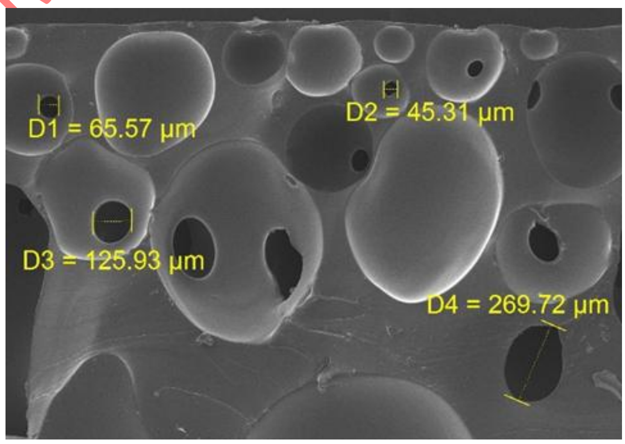

Fig. 2. SEM micrograph showing the microporous surface of the folate-polymer composite sample (500 μm scale bar).

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The biocompatibility of the composite is due to the proximity of the chemical composition of the urethane group - CO-NH - polyurethane to the peptide group of proteins. Study of the histotoxicity was performed by the cell culture assay, using subcutaneous cellular tissue of the white lab rats according to the method of quantitative study of the fibroblasts development at the cell culture [14].

The process of biodestruction of the adhesive composition occurs as a result of a combination of non-enzymatic hydrolysis and cellular resorption of the polymer by two main groups of cells: macrophages that phagocyte microparticles of polymer and gigantic cells of foreign bodies that penetrate the implant through lysis and divide it into fragments. The products of biodestruction are gradually removed from the body through the urinary system and the gastrointestinal tract without accumulation in the liver and kidneys, as radioisotope analysis show [15, 16].

Folic acid (vitamin B9, N-pteroyl-L-glutamic acid) is known as a substance that exhibits its biological activity due to the presence of the inner-ring cycle by joining hydrogen atoms to carbon atoms and nitrogen to form tetrahydrofolic acid, which performs the biochemical function of coenzyme in intermolecular transport of single-carbon groups of different degrees of oxidation, which is very important for the further synthesis of nucleic acids (RNA and DNA) [17].

Some scientists point to the improvement in the gum's protective abilities and, respectively, reducing inflammation and even phenytoin-induced gingival overgrowth, especially when patient received an oral supplementation of 5 mg of folic acid daily [18].

The study of biocompatibility and bioactivity of folate-containing composites was performed on 36 white laboratory rats weighing 150-200 g, who were implanted with polymer samples measuring 10x10x4 mm. The observation time was 7, 14 and 30 days.

RESULTS:

To date, the physical and mechanical properties of composite materials with different percentages of folic acid - FA (0.248% and 0.48%) and without it – have been studied. The obtained data indicate that the experimental samples with FA had greater adhesion and strength characteristics compared to the control samples.

Data of the infrared spectroscopic studies indicate that FA immobilization on polymeric sample does not change the chemical structure of the composition, and the

FA itself binds to the adhesive basis, mainly due to the formation of hydrogen bonds, which are more easily destroyed under the influence of the environment of the organism.

Morpho-histological studies have shown that FA in the composite material inhibited the exudative phase of inflammation. The restriction of exudation resulted in the slow release of the formed blood elements and plasma proteins, which ultimately affected the course of the reparative process in the surrounding tissues, contributing to the faster formation of a connective tissue capsule, characterized by a slight thickness and high degree of maturity. Round-cell infiltration of surrounding tissues also decreased around experimental FA samples.

The investigated composite materials are biocompatible with tissues of experimental animals, do not cause chronic inflammatory reaction, retain their structure during implantation for a long time (up to 30 days). After 30 days there is a gradual germination of the connective tissue grafts deep into the polymeric implant.

Morphological studies have shown that the changes that occur after implantation of composite materials correspond to the picture of normal healing of trauma and is a physiological response of a living organism to the presence of a foreign body.

Starting from the 14th day of the study, a rather thin and mature connective tissue capsule is defined around all implanted samples. It is a dense, unformed connective tissue, the bulk of which consists of fibrous structures – collagen fibers. Between the beams of collagen fibers there are somewhat elongated and oval cells of a fibroblast tissue of varying degrees of maturity, which are larger in diameter, mainly directed along fibrous structures. In addition, there are leukocytes, lymphocytes and macrophages in different numbers.

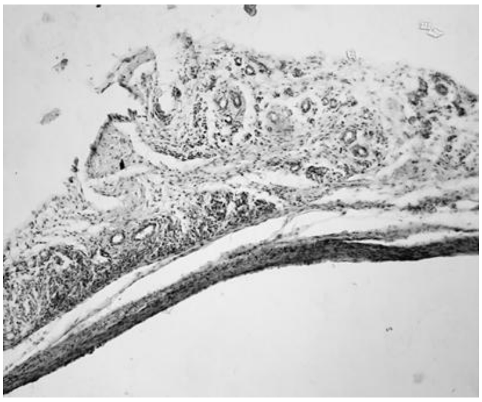

Fig. 3. A large number of blood vessels with normal microcirculation around the sample from 0,248% FA on the day 14 of the experiment Hematoxylin and eosin staining. X120

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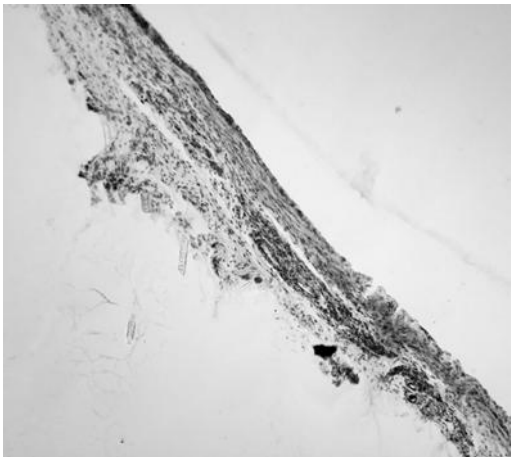

Fig. 4. Thin connective tissue capsule around the sample with 0,248% FA after 30 days after implantation. Hematoxylin and eosin staining. X120

It should be noted that the intensity of cellular reactions, in particular, circular cell elements, around the composite material of the composition with 0.248% FA was the lowest. Implantation of this composite material caused only a local cellular response aimed at the implementation of protective and compensatory mechanisms of the body in response to the presence of the foreign body [19].

Experimental studies of bone regeneration in 36 white rats have shown that this process occurs both at the edges of the bone defect and through the transformation of the connective tissue that germinates intensively in the pores of the polymer composition.

According to preliminary histological studies [20], duration of replacement of a polymeric composition by a bone takes place within 6-8 months (Fig. 5).

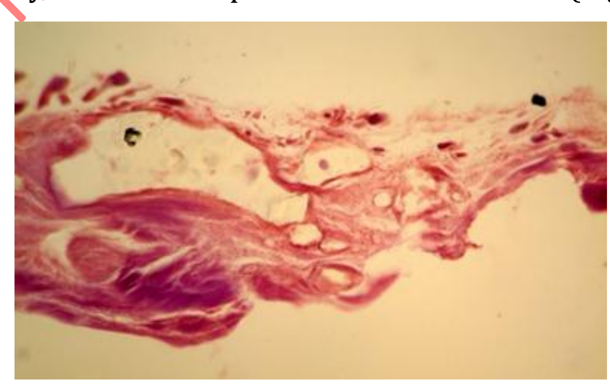

Fig. 5. Deep sprouting of connective tissue in the pores of polymeric material after 6 months of implantation.

Hematoxylin and eosin staining. X120

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For clinical use, the adhesive base, filler (folic acid) and polymerization accelerator are successively added into a sterile syringe in the above order, where mixing is carried out. Then using the cannula "Medical glue" is injected into pre-dried bone defect, tightly clamping the tissue. The glue is applied after carrying out all necessary surgical manipulations, using modern tooth-preserving techniques and microsurgical techniques [21]. The primary bonding between adhesive material and tissue surface requires at least 5 minutes, after that time the surgical wounds are sewn.

Biodegradation of the polymer composites occurs at the body in a parallel way with bone formation, that provide strength of contact of the implant and bone and postoperative teeth stability, which roots protruding to the cavity of periradicular bone defects.

Conclusions.

1. The prolonged biological effect, biocompatibility, gradual biodegradation, adhesion strength properties of folate-polymer composites have been experimentally confirmed; Such composites can be used in clinical practice for the purpose of surgical treatment of periradicular bone defects.
2. Properties of polymeric samples allow simultaneous performance of structural and barrier functions, which is relevant at the time of applying the technique of guided bone regeneration.
3. An important component of successful and prospective treatment of periradicular bone defects is the combination of biologically justified concepts and techniques with the clinical application of modern equipment, tools and materials.
4. Folate-polymer composites requires further laboratory, clinical, roentgenological and statistical studies.

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