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Studying Clinical And Pathomorphological Changes In Gingival Tissues When Applying Platelet-Rich Autoplasma Regenerative Method.

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#### **ABSTRACT**

The problem of high incidence and treatment need of the periodontium diseases is one of the priorities for the modern dental practice. However, despite the wide range of drugs that affect both the microflora of periodontal pockets and the various pathogenesis mechanisms of the inflammatory process, no significant reduction in the number of patients with periodontal diseases has been observed. In this regard, the search for new methods and means of enhancing the effectiveness of therapeutic effect on the pathological focus of inflammation in the periodontium is in process. The method of choice in the dental practice can be a drug-free method such as autohemotherapy. The use of platelet-rich plasma is today one of the few opportunities to modulate and enhance wound healing process and resist an infection without the use of drugs.

Keywords: periodontal tissue, autohemotherapy, inflammation, treatment

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#### INTRODUCTION

Tissue regeneration is an urgent matter of dentistry. This is due to the prevalence of periodontal disease, high requirements of patients to the ensured treatment after surgery, as well as the patients' permanent desire to have a beautiful smile. The prevalence of inflammatory periodontal diseases among the adult population in the world remains at a high level and shows no tendency to decrease [9].

Treatment of periodontal disease requires not only to remove inflammation, but also to prevent further bone loss, restore color, shape and structure of the gum tissue for a longer period, to stimulate regeneration processes and thereby reduce the frequency of exacerbations [2].

Today, drugs for the treatment of periodontal disease include the application of general and local medicinal products [5]. All that is administered orally is either antibacterials that may also be local, or osseostimulating drugs. Antibacterials only remove inflammation and do not launch the regeneration processes. All osseostimulating drugs are primarily not the cheapest one, and require long-term use, and hence, the corresponding control over the state of kidneys and gall bladder. Therefore, there is a need for additional time and patient's money for check-up analysis [8, 10]. Topically, drugs for treatment of periodontal and oral mucous diseases can be either applicable or injectable [4]. It has been repeatedly shown that the absorbance of applicational drugs is only 7.10%, so the efficiency of these methods is extremely low. Injectable drugs are often only temporarily remove inflammation and improve hemodynamics, and do not directly start the regeneration processes. Nevertheless, one must remember that the tissues of this region is already structurally destroyed, and thus the supply of nutrients and the "construction" of cells is difficult, and the resulting effect will be temporal and poorly pronounced. Development of a large number of new highly effective medicines is due to the achievements of modern medicine in the prevention and treatment of many serious diseases in both general and dental practice [7]. Along with success new challenges have occured: the higher the activity of the drug is, the more side effects it has [11]. A significant increase in the number of complications from the medication raises concerns of both doctors and patient, which provokes the search for alternative methods of treatment [1,6].

Last decade is characterized by a significantly grown interest in the use of platelet autoplasma. Platelet-rich autoplasma has a number of useful properties such as acceleration of tissue regeneration, antiinflammatory effect, and reduction of pain syndrome. Nowadays, a autohemotherapy is actively used in surgery, traumatology, orthopedics, sports medicine, cosmetology, and dermatology. Currently, there is information occurring in the professional literature about the use of platelet autoplasma in treating the inflammatory diseases of the maxillofacial region. Its applicability in dentistry is under consideration, based on the activation of human functional reserves reduced under the influence of either adverse environmental factors or disease [3, 12]. However, clinical studies based on demonstrative data are scarce, and the obtained results require further study.

#### **OBJECTIVE OF RESEARCH**

To study the efficiency of autohemotherapy in the treatment of moderate chronic generalized periodontitis.

# **MATERIALS AND RESEARCH METHODS**

The clinical part of the study involved examination and treatment of 90 patients with moderate chronic generalized periodontitis, divided into 2 equal groups: group I (control) - a conventional therapy; group II - treatment with the use of platelet autoplasma. All patients received a comprehensive treatment.

The patient was examined by standard methods (basic and additional): survey, visual examination, index indicators of the state of hard dental tissues and periodontal tissues, and X-ray examination.

Clinical observation was performed in all groups of patients during the first visit, on day 7, and after 3, 6, and 12 months. All information was recorded in the case records of dental patients.

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Platelet-derived autoplasma was prepared from the patient's own blood with further centrifugation and injection of finished substance in the affected area. Treatment was conducted in courses. One course consisted of 3 procedures (1 vial per visit) with an interval of 7 days. The obtained platelet autoplasma (about 3.5 ml) was administered to the target area. When using this method in periodontics: 0.1-0.2 ml to marginal gingiva area per 3.2 mm², 0.3-0.5 ml into transitional fold per one dentition segment consisting of 1-2 teeth. Injection in the marginal gingiva was performed both from vestibular and from oral side.

The experimental part involves the study of the features of morphological changes in periodontal tissues in experimental animals under the influence of platelet autoplasma. Eight dogs with moderate chronic generalized periodontitis, which were divided into groups depending on the type of treatment, were used as the object of study of PRP-therapy influence on periodontal disease. All animals underwent professional oral hygiene. Group I (control) - a conventional therapy without platelet autoplasma; group II - treatment with the use of platelet autoplasma.

The experimental dogs had their blood drawing from the vein, in the amount of 7 ml. Blood was sampled by the standard method using a tourniquet, alcohol wipes, a needle, a test-tube holder adapter, plaster, and specialized test-tubes "PlasmoliphtingTM". After sampling the blood, a test tube was placed into a centrifuge "EVA 20", at 3200 rpm for 5 minutes. Using special test tubes "PlasmolifhtingTM" allowed us to obtain 1.5±0.5 ml of platelet plasma; in addition, a separating gel ensures high-quality filtration of plasma and good fixation of the erythrocyte clot.

Periodontal status was assessed by conventional techniques used in dentistry to detect signs of inflammation in the periodontium: Schiller-Pisarev test (presence of gingival staining), identification of gum congestion and swelling, and the PMA index. To eliminate the influence of additional factors, related to individual characteristics of the laboratory animals, on the final result of the experiment, the observational groups were formed of the same animal species. Clinical observation of periodontal state of the animals was performed every day throughout a week. The study was conducted in compliace with the rules of good laboratory practice when conducting preclinical studies in the Russian Federation (GOST R 50258-92) and the rules of humane treatment of animals (Report of the AVMA Panel on Euthanasia JAVMA, 2001), as well as the rules of the International recommendations of the European Convention for the protection of Vertebrate Animals used for experimental studies (1997). The experiments were conducted with the permission of the Research Ethics Review Committee of Volgograd State Medical University (protocol No. 214 - 2015 of 04.29.2015).

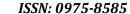
Data obtained from studies were processed by variation statistical method on the IBM PC/AT Pentium-IV Windows 2000 using the application package Statistica 6 (Statsoft-Russia, 1999) and Microsoft Exsel Windows 2000. Statistical analysis was performed by variation statistical method with defining the average value (M), its mean error ( $\pm$  m), and evaluating the significance of difference in the groups using Student's t test (t). The difference between the compared indicators was considered significant at p<0.05, t≥2.

### **RESULTS**

All dogs included in the experiment had dental plaque, bleeding, redness of papillary and marginal gingiva revealed at the time of the examination. At the time of the study, PMA index values in the groups were as follows: group I - 32.3±0.953 points, group II - 32.29±0.967 points. The Muhlemann index values in the groups were as follows: group I - 1.552±0.287 points, group II - 1.542±0.268 points.

After professional oral hygiene, which included removal of dental deposits, dental polishing with brush and "Depural" paste, and the gum irrigation with antiseptic of 0.05% bigluconate chlorhexidine, the dogs were divided into two groups, according to the previously selected treatment method. The conducted treatment resulted in objective improvement: normalization of gum color and lack of edema. PMA index decreased in group II up to 28.74±0.982 points, in the control group - up to 28.81±0.808 points. Bleeding index decreased in group II up to 0.458±0.195 c.u., in control group - up to 0.529±0.199 c.u. Clinical pattern after 2 weeks of treatment did not change significantly in the control group, while the group treated with platelet autoplasma showed complete normalization of the status of periodontal tissues: pale pink gingiva tightly covered the neck of the teeth.

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It was noted that in group II treated with platelet autoplasma, the first signs of inflammation elimination such as normalization of color, configuration of the interdental papillae and marginal gingiva, as well as a significant decrease (p<0.05) in gum bleeding were already observed on the second day. The signs of inflammation in the control group disappeared 2-2.5 times slower (p<0.05).

Thus, the assessment of the clinical periodontal status after treatment of chronic generalized periodontitis in experimental animals revealed that the resolution of the inflammatory processes occurred usually on day 3. However, the application of platelet autoplasma allowed reducing the treatment time up to 2 days in comparison to the control group.

Morphological assessment of the efficacy of platelet autoplasma used in dogs' gums for the treatment of periodontal disease has shown atrophy of stratified squamous non-keratinizing epithelium, focal fibrosis (sclerosis) of the underlying tissue, 3 days after the experiment, in the control group, where the defect was healed without additional treatment, while the group II showed the groups of newly formed vessels, and the presence of lymphohisticytic diffuse infiltration with admixture of neutrophils in the underlying tissue. On day 21 of the experiment, group II shows decrease in the number of inflammatory cells in the tissues with more localized arrangement in tissues, while the number of newly formed cells - immature fibroblasts and collagen - increases inside the infiltrates. On day 28, we could observe the normally structured gums.

The control group on day 21 showed the reduction in the number of inflammatory cells, and the phenomena of focal acanthosis and parakeratosis of the stratified squamous non-keratinizing epithelium. On day 28, a fragment of mucosa lined with hyperplastic, multilayered, non-squamous epithelium with signs of acanthosis, the underlying tissue with diffuse lymphohistiocytic infiltration with a mixture of neutrophils were revealed. That is, reparative processes had just started.

As we can see from clinical data, in some cases the first therapeutic effect can be observed in the oral cavities of the patients of group II after a week, which is manifested by improved hygienic and periodontal indices, decreased gum bleeding and hyperemia, and the recovered physiological color of the gum. Some patients had no complaints, against the positive dynamics of diagnostic methods indicators (PMA was 16.9±1.7; Muhlemann gum bleeding index - 1.29±0.1 c.u.). Data obtained in the experimental group differed significantly from the data obtained during examination of the control patients (PMA - 36.5±1.0; bleeding index - 2.1±0.3).

3 months after treatment, the stabilization of the pathological process can be clinically confirmed in the group II. Clinical stabilization of inflammatory process in periodontitis was characterized by the absence of bleeding, swelling, redness of gums, normalization of gingival density and contour, reduced probing depth and tooth mobility, and the persistent level of the epithelial attachment.

After 6 months, we obtained the following clinical and radiographic findings:

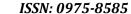
- no patient's complaints;
- clinically normal periodontal tissues;
- radiologically normal periodontal tissues; the absence of osteoporosis, normalization of bone looping of the interdental septa.

#### After 12 months:

- no patient's complaints;
- achieved clinical stabilization of the inflammatory process in the periodontium;
- identified radiological signs of stabilization of the inflammatory process in the periodontium;
- no growth of indicators of periodontal indices;
- restoration of dental function, diction, esthetics.

At the same time, the inflammation phenomena in the control group were stopped 2.5 times slower than in the experimental group.

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#### **SUMMARY**

Results of the study confirmed the clinical efficacy of platelet autoplasma in the treatment of generalized periodontitis. The application of autohemotherapy in the comprehensive treatment of periodontal diseases allows stopping inflammation in the periodontium at an earlier date as compared to the conventional treatment scheme. The dynamic of the PMA index values was characterized by a significant decrease in all groups, but a bit greater reduction by 24% in this index as compared to its value before the treatment was observed in the experimental group (from 32.61±0.96 points to 16.9±1.7 points).

The Muhlemann bleeding index decreased in all groups, but the experimental group showed its more pronounced decrease from the initial value by 82%, while the control group had the same reducing by 65%.

As can be seen from the presented results, the reduction of swelling, redness, bleeding and the periodontal indices in group II treated with platelet autoplasma was more pronounced, as evidenced by significant difference in the indicators.

Comprehensive treatment of patients with the use of Plasmolifting technique has ensured faster, more intensive than under conventional medical treatment normalization of inflamation parameters. The group II showed more pronounced positive dynamics throughout the study as compared to the group I, where autochemotherapy was excluded from treatment.

#### CONCLUSION

The findings of clinical study of the platelet-rich plasma efficiency in the therapeutic treatment demonstrate convincingly the accelerated reduction of the inflammatory process.

#### **CONFLICT OF INTERESTS**

The author declares that the provided information has no conflicts of interest.

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