The paper presents the results of pathogenetic rationale for the use of gel with lipopolysaccharide for gingivitis modelling. It shows, that lipopolysaccharide applications lead to significant increase in activity of elastase by 26.8%, malonic dialdehyde by 32.8%, urease by 78.9%, degree of dysbiosis by 32.6%, decrease in activity of lysozyme by 57.6% and antioxidant-prooxidant index by 29.7%. Thus, lipopolysaccharide application leads to the progression of inflammation processes in gums of experimental animals, dysbiosis and oxidative stress, that is pathogenetic links of gingivitis progression in humans as well.

**Key words:** gingivitis, periodontal disease, experimental model, experimental animals, lipopolysaccharide

Periodontal diseases continue to occupy a special place among all dental pathology. This is due to a number of reasons, among which the main are the diversity of nosological forms, etiological factors and pathogenetic mechanisms, the lack of highly effective means for their prevention and treatment. Those periodontal diseases that arise in the setting of the pathology of various organs and systems, particularly the digestive system, occupy a special place [4, 7, 9].

A significant role in the progression of dental pathology in general and periodontal diseases, in particular, belongs to the liver – a human internal organ, which functioning is associated with the performance of such vital functions as antitoxic, regulatory, metabolic [2, 10, 11].
In the last decade, attention of researchers is attracted to such aspects of the pathogenesis of inflammatory and inflammatory-dystrophic periodontal diseases as free-radical lipid oxidation, changes in the immune status, the state of protective periodontal enzyme systems [3,8].

It is the modeling of the pathological process in periodontal tissues that contributes to the in-depth study of gum diseases pathogenesis, the development and confirmation of the effectiveness of drugs use and non-drug treatment methods of this pathology. Thus, when modelling hepatitis in rats by introducing per os an oily solution of carbon tetrachloride (CCl4), the progression of degenerative changes in the epithelial layer of the oral mucosa and gums with a decrease in RNA content in epithelial cells was shown. The proposed method also led to a significant increase in total proteolytic activity, in the activity of alkaline phosphatase, and an increase in the concentration of the lipid peroxidation marker - malonic dialdehyde. [8].

Modeling hepatocohelectisitis in rats by introducing per os tuberculostatics (isoniazid + rifampicin + pyrazinamide) during 2 weeks led to periodontal tissues atrophy, increased gum inflammation markers level (malonic dialdehyde, total proteolytic activity, acidic phosphatase), and significantly reduced an antioxidant protection level (decrease in catalase activity) [1].

One of the toxic factors of hepatogenic origin is lipopolysaccharide (LPS), which forms during cell walls hydrolysis of pathogenic and conditionally pathogenic gram-negative bacteria species. Inducing immune defense factors stimulation, LPS triggers a range of pro-inflammatory and immunochemical reactions [12, 14]. In a healthy body liver neutralizes almost all LPS coming from the intestine. However, with liver function abnormality LPS enters the systemic circulation and exerts its pathogenic effect on many organs and tissues [12]. Part of LPS can also form in the oral cavity tissues due to oral gram-negative bacteria [13].

In view of the foregoing, it is relevant to assess the role of LPS as one of the factors of hepatogenic origin in the periodontal diseases progression.  

The purpose the study was pathogenetic rationale for the model of chronic gingivitis with use of LPS.

Materials and methods. For an experimental study of the LPS effect 14 male WAG rats were used with an average weight of 380 ± 14 g. Half of the experimental animals (species) served as a control, and the remaining 7 males (main group) were once applied 0.5 ml of LPS gel containing 50 mg / ml (in terms of 1 kg of body weight) on the gums. The gel was applied 30 minutes before meals. On the second day, the animals were removed from the experiment, the gums were dissected out and standard biochemical parameters of inflammation (elastase and MDA), dysbiosis and antioxidant protection in the homogenate were determined. The inflammatory response was evaluated by the activity of such markers as malonic dialdehyde (MDA), elastase and catalase. The antioxidant prooxidant index API was calculated by the catalase activity ratio and MDA concentration. The dysbiosis degree was judged by the ratio of microbial contamination level (urease) and lysozyme activity [5, 6].

All the manipulations with experimental animals were carried out in accordance with the provisions of the European Convention for the Protection of Vertebrate Animals, which are used for research and other scientific purposes (Strasbourg, 1986), the Law of Ukraine “On the protection of animals from cruel behavior” (2006).

Table 1

<table>
<thead>
<tr>
<th>Indicators</th>
<th>Control group, n=7</th>
<th>Main group, n=7</th>
</tr>
</thead>
<tbody>
<tr>
<td>Elastase, mkat/kg</td>
<td>41±2</td>
<td>52±2 p=0.0000</td>
</tr>
<tr>
<td>MDA, mmol/kg</td>
<td>13.7±0.9</td>
<td>18.2±1.0 p=0.0000</td>
</tr>
<tr>
<td>Urease, mkat/kg</td>
<td>1.28±0.14</td>
<td>2.29±0.28 p=0.0000</td>
</tr>
<tr>
<td>Lysozyme, u/kg</td>
<td>393±70</td>
<td>167±13 p=0.0000</td>
</tr>
<tr>
<td>Catalase, mkat/kg</td>
<td>5.84±0.17</td>
<td>5.46±0.16 p=0.0010</td>
</tr>
<tr>
<td>API, units</td>
<td>4.25±0.35</td>
<td>2.99±0.27 p=0.0000</td>
</tr>
<tr>
<td>Dysbiosis degree, units</td>
<td>1.00±0.10</td>
<td>4.26±0.50 p=0.0000</td>
</tr>
</tbody>
</table>

Note: p - significance level of differences with the control group (t-test for independent samples)

The statistical processing of the results was performed using the program Statistica 6.0 (Statsoft Inc., USA). The arithmetic mean (M) and the average error of the arithmetic mean value (m) were calculated. The estimation of the probability of the average values (p) difference was executed using Student’s t-test. The nature of the relationship between the ranking structures was determined using the Spearman correlation coefficient (ps).

Results and their discussion. Conducted biochemical studies of gums homogenates of experimental animals showed that, compared with control animals, the elastase level significantly increased by 26.8% (p = 0.0000), MDA by 32.8% (p = 0.0000), urease by 78.9% (p = 0.0000), degree of dysbiosis by 326% (p = 0.0000), reduction of lysozyme content by 57.6% (p = 0.0000) and API by 29.7% (p = 0.0000) (table 1). In addition, catalase reducing trend by 6.5% (p = 0.0010) was marked.
The findings indicate progression of inflammation in the gums of animals, dysbiosis, and anti/prooxidant balance disorder.

Determining the degree of deviation from the standard values of the studied parameters found that a decrease in the activity of lysozyme possessed the most pronounced deviance (t = 11.3; p < 0.001) (fig. 1). The second rank belongs to a moderate increase in the degree of dysbiosis. (t = 5.92; p < 0.001). The following are indicators with minor deviations from the standard. In order of rank significance they are: increased elastase (t = 3.57; p < 0.01 – the third rank), MDA (t = 3.0; p < 0.05 – the fourth rank), urease (t = 2.94; p < 0.05 – the fifth rank) and reduction of API (t = 2.6; p < 0.05 – the sixth rank). The last rank position belongs to the tendency to reduce the level of catalase (t = 1.48; p < 0.05) (fig. 1).

It should be noted that the direction of listed above deviations of the values of all indicators has a pathogenetic significance.

As for the degree of individual links of pathogenesis involvement in the pathological process (fig. 2), dysbiosis manifestation is most marked (t = 7.0; p < 0.001). The second important role belongs to the inflammation phenomena (t = 3.57; p < 0.01), and the last rank is occupied by oxidative stress, that is, the degree of oxidative and antioxidant mechanisms imbalance in the processes of free radical oxidation (t = 2.24; p < 0.05).

Thus, LPS causes inflammation in the gums of experimental animals. The greatest pathogenetic significance in periodontal diseases progression when using a gel with LPS belongs to dysbiosis, the least – to anti-prooxidant system state.

Modeling of the pathological process in periodontal tissues using oral LPS gel leads to a sharp activation of inflammatory markers, a decrease in the antioxidative protection level, which is consistent with the data obtained by Bykov E.M. et al., Sirak S.V. [3, 8]. At the same time, the suggested method of the gingivitis experimental reproduction permits, unlike other models [1], to reduce the degree of local destructive influence on the gum epithelium and to perform a profound study of the inflammatory process pathogenetic links in the periodontium, based on the dysbiosis.

**Conclusion**

The results obtained substantiate pathogenetically the use of oral LPS gel as an experimental model of gingivitis. The proposed option allows to quickly and with minimal cost reproduce the inflammatory process in the gums of rats, the most appropriate for the clinical manifestation of gingivitis in humans.

**References**

Наведено результати патогенетичного обґрунтування застосування гелю з ліпополісахаридом для моделювання гінгівіту. Показано, що аппликація ліпополісахариду призводить до достовірного підвищення активності еластази на 26,8%, малонового діальдегіду на 32,8%, у рецесії на 78,9%, стійкість дисбіозу на 32,6%, зниження активності лізому на 57,6% та антиоксидантно-проксидантного індексу на 29,7%.

Ключові слова: гінгівіт, захворювання пародонта, експериментальна модель, експериментальні тварини, ліпополісахарид.

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МОРОФОФУНКЦІОНАЛЬНИЙ СТАН ГЕМОМІКРОЦИРКУЛЯТОРНОГО РУСЛА СІТКІВКИ ПІСЛЯ ОДНОРАЗОВОГО ВВЕДЕННЯ КРИЮКОНСЕРВОВАНОЇ ПЛАЦЕНТИ НА ТЛІ ГОСТРОГО ЕКСПЕРИМЕНТАЛЬНОГО АСЕПТИЧНОГО РЕТИНІТУ У ЩУРІВ

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Робота є фрагментом ЦДР «Експериментально-морфологічне вивчення дії трансплантацій криюконсервованої плacentи на морфофункціональний стан ряду внутрішніх органів», № державної реєстрації 0113У006185.

На початку минулого століття професор В.А. Філатов обґрунтував метод тканинної терапії [1,2,5], завдяки цьому винаходу тканина терапія отримала широкий розвиток і клінічне застосування в офтальмології.

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