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COMPREHENSIVE BIOCHEMICAL DETERMINATION OF THE THERAPEUTIC AND PREVENTIVE EFFECTIVENESS OF HERBAL REMEDIES FOR PERIODONTAL DAMAGE IN RATS THAT CONSUMED FRIED SUNFLOWER OIL

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Abstract

Background. Periodontitis is one of the most common human diseases (more than 80% of the adult population of highly developed countries). In the pathogenesis of periodontitis, osteoporosis of the periodontal bone tissue plays a decisive role, which occurs as a result of a significant decrease in mechanical load on the jaws, a deficiency of osteostimulating factors (vitamin D, calcium, omega-3 polyunsaturated fatty acids, sex hormones), as well as as a result of the development of oral dysbiosis. To investigate the therapeutic and prophylactic effectiveness of multifunctional phytomedicines using a complex biochemical method based on the total determination of the degree of changes in the level of several biochemical markers of pathology and protection.

Methods. Fried sunflower oil (FSO) was obtained by thermal peroxidation of ordinary

unrefined sunflower oil at a temperature of 180°C in the presence of a catalyst. Biological studies were conducted on white Wistar rats, which consumed a mixed feed containing 2 g/kg of FSO for 75 days. As phytopreparations, we used multifunctional products developed by us containing bioflavonoids, lysozyme, mint and amaranth seed extracts, as well as prebiotics and calcium citrate. Comparison phytopreparations («Kvertuline» and «Lysozyme-forte») were administered with feed at a dose of 300 mg/kg, starting from the 31st day of the experiment. One group of rats that consumed FSO received oral irrigation of the oral cavity with 2 ml of the «Mint + Amaranth Extract» dental elixir. Rats were euthanized, gums and alveolar process of the mandible were isolated. In the gum homogenates, biochemical markers of pathology were determined, namely the activity of elastase, urease and the content of malondialdehyde (MDA), as well as the level of biochemical markers of defense systems, namely the activity of lysozyme, catalase and the antioxidant-prooxidant index API was calculated. In the homogenate of bone tissue, the activity of acid (AP) and alkaline (ALP) phosphatases was determined, the calcium content was determined, and the mineralization index (MI) was calculated based on the AP/ALP ratio. The degree (in %) of the decrease in the level of pathology indicators (elastase, urease, MDA, acid phosphatase) and the increase in the level of protection markers (lysozyme, catalase, API, alkaline phosphatase, calcium, MI) was calculated, and based on these data, the therapeutic and prophylactic efficacy (TPE) of each herbal preparation was calculated.

Results. It was found that the consumption of FSO increases the activity of elastase in the gums by 55.2 %, urease by 26 % and MDA content by 72.8 %, but reduces the activity of lysozyme by 27.7 %, catalase activity by 15.8 % and the antioxidant-prooxidant index API by 51.3 %. In the periodontal bone tissue, the consumption of FSO increases the activity of AP by 206.5 %, but reduces the activity of ALP by 23.6 %, the mineralizing index MI by 61.5 % and the calcium content by 20.3 %. In rats that consumed FSO on the background of herbal preparations, the activity of elastase, urease and MDA content increases significantly less and the activity of lysozyme, catalase and API index decreases significantly less. In periodontal bone tissue, when consuming FSO against the background of herbal remedies, the activity of AP increases significantly less (by 4 – 10 times), the activity of ALP decreases by 3 – 10 times less, and the MI index decreases by 1.5 – 3 times less. Consumption of FSO against the background of herbal remedies reduces the calcium content by 2 times less.

Conclusion. The therapeutic and prophylactic efficacy (TPE) calculated according to these indicators in the gums was the highest (76.5 %) in the dental elixir. In the bone tissue, TPE was also the highest in the dental elixir.

Keywords: periodontitis; fried oil; herbal medicines; methods for assessing

therapeutic and prophylactic effectiveness.

Introduction

Periodontitis (inflammatory and dystrophic process in the periodontal tissues) is one of the most widespread diseases of people (more than 80% of the adult population) [1, 2], and recently even children have been suffering from periodontitis [3]. The causes of periodontitis can be various factors: genetic, microbiological, neuro-endocrine, bad habits, etc., but the main reason for the development of periodontitis is the non-physiological nature of our diet. What does it consist of? First of all, the nature of nutrition has changed significantly: we eat less and less solid food and more and more soft, mashed food. As a result, the mechanical load on the jaw bone tissue is significantly reduced and this leads to the development of oral osteoporosis [4]. Osteoporosis of the periodontal bone tissue creates all the conditions for the formation of periodontal pockets, in which bacterial microflora develops, including periodontal pathogenic, which causes the development of the inflammatory process in the periodontium [5, 6].

Secondly, not only the physical state of our food has changed, but also the composition of our diet, in which most dishes undergo heat treatment, and in many cases high-temperature (more than 100 ºC). Dishes obtained as a result of frying (temperature more than 200 ºC) have become extremely popular. High-temperature processing of food products leads to the destruction of substances useful for the body (vitamins, amino acids, fatty acids) and to the formation of thermal peroxidation products harmful to the body (antimetabolites, lipid peroxides, various aldehydes and ketones, trans-isomers of fatty acids, etc.) [7]. Constant consumption of heat-treated foods leads to intoxication of the body, especially of organs such as the colon mucosa and the liver [7]. The function of almost all organs and systems of the body, including the state of the periodontium (hepato-oral syndrome [8], depends on the state of the liver.

A significant nutritional problem is excessive fat consumption (more than 20 % of calories), especially with a high content of fatty acids such as palmitic and linoleic (palm, ordinary sunflower, soybean, corn, rapeseed oils) [9]. We have found that linoleic acid (the main component of ordinary sunflower oil) inhibits the endogenous biosynthesis of omega-3 polyunsaturated fatty acids, and these acids stimulate osteogenesis and prevent the development of osteoporosis [10].

The third reason for the development of periodontitis is a violation of the nature of the interaction between the macroorganism and the endogenous microbiota, which is a component, functionally active part of our body [11]. This primarily concerns the oral cavity, in which, due

to improper oral hygiene, dysbacteriosis (dysbiosis) usually develops due to a significant decrease in the number of probiotic microflora. As a result, conditions are created for the development of conditionally pathogenic microbiota, which causes the activation of inflammatory and dystrophic processes in the periodontium.

The fourth cause of periodontitis is stress, in particular its third post-stress reaction, in which the level of corticosteroids increases, which also cause osteoporosis [12].

The aim of the study

To investigate the therapeutic and prophylactic effectiveness of multifunctional phytomedicines using a complex biochemical method based on the total determination of the degree of changes in the level of several biochemical markers of pathology and protection.

Materials and research methods

The experimental part of the study was conducted in full accordance with our previous work on determining the TPE of herbal preparations on the liver condition of rats receiving FSO [13].

The characteristics of the herbal preparations used in this work are presented in Table 1.

Table 1. Characteristics of herbal medicines

Phytopreparation	Composition	Regulatory documentation
«Kvertuline» (dietary supplement)	Quercetin, inulin, calcium citrate	TC 10.8-13903778-040:2012
«Lysozyme-forte» (dietary supplement)	Lysozyme, quercetin, inulin, calcium citrate, hesperidin, skim milk powder, ascorbic acid	TC U 10.8-37420386-004:2016
Dental elixir «Mint + amaranth extract» (hygienic product)	Mint extract, amaranth extract, inulin, cetavlon, citric acid, hesperidin	TC U 569A-13903778-001-92

The dose for bulk preparations («Kvertuline» and «Lysozyme-forte») was 300 mg/kg of live weight of rats, and the dental elixir was administered into the oral cavity of rats in an amount of 2 ml. The administration of herbal preparations began on the 31st day of the experiment and was completed on the 75th day.

The periodontal condition in rats receiving FSO or FSO on the background of herbal preparations was assessed by the level of biochemical markers: in the gums, by the level of activity of elastase, urease, lysozyme, catalase, by the content of malondialdehyde (MDA) and by the level of antioxidant-prooxidant index API.

In the periodontal bone tissue (alveolar process of the mandible), the activity of acid phosphatase (AP), alkaline phosphatase (ALP), calcium content, and the mineralizing index MI were determined by the AP/ALP ratio (Levitsky et al., 2005).

The state of gum pathology was assessed by the sum of the indicators of increased levels of elastase, urease, MDA and the sum of the indicators of decreased levels of lysozyme, catalase and API.

The state of periodontal bone pathology was assessed by the sum of the following indicators: increased AP activity, decreased ALP activity, decreased calcium content, and decreased MI index.

Statistical processing of the research results was carried out in accordance with the instructions of the computer program Statistica 7.0. The reliability of the results in the comparison groups was carried out using the Student's t-test.

Results and discussion

Table 2 presents the results of determining the level of biochemical markers of pathology in the gums, namely the activity of elastase, urease and MDA content. In rats that consumed FSO, the activity of elastase and MDA content significantly increased.

Table 2. The effect of herbal medicines on the level of biochemical markers of pathogenicity in the gums of rats fed fried sunflower oil (FSO))

№№	Group	Pathogenicity markers		
		Elastase, mcat/kg	MDA, mmol/kg	Urease, micro-cat/kg
1	Control	37,3±3,5	18,4±1,5	0,73±0,12
2	FSO	57,9±1,6 $p<0,01$	31,8±1,6 $p<0,01$	0,92±0,06 $p>0,05$
3	FSO+ «Kvertulin»	48,9±2,2 $p<0,05$; $p_1<0,01$	26,1±1,2 $p<0,05$; $p_1<0,05$	0,77±0,07 $p>0,05$; $p_1>0,05$
4	FSO + «Lysozyme-forte»	49,4±1,7 $p<0,05$; $p_1<0,05$	24,9±1,1 $p<0,05$; $p_1<0,01$	0,78±0,07 $p>0,1$; $p_1>0,05$
5	FSO + dental elixir «Mint + amaranth extract»	41,6±2,4 $p>0,05$; $p_1<0,01$	22,3±1,8 $p>0,05$; $p_1<0,01$	0,75±0,11 $p>0,2$; $p_1<0,05$

p – compared to the group 1; p_1 – compared to the group 2.

Urease activity in the gums of rats treated with FSO shows only a tendency to increase. All herbal preparations significantly reduced the activity of elastase and MDA content. Urease activity in the gums of rats that received herbal remedies on the background of FSO shows a tendency to decrease.

Table 3 presents the results of determining the level of biochemical markers of protection in the gums, namely the activity of lysozyme, catalase and the level of the API index. The activity of lysozyme and catalase in the gums of rats that received herbal preparations on the background of FSO is normalized. As for the API index, it significantly increases in the gums of rats that received herbal preparations on the background of FSO, and in the group of rats that received applications with dental elixir, this indicator increases to the control level.

Table 3. The effect of herbal preparations on the level of biochemical markers of defense systems in the gums of rats receiving fried sunflower oil (FSO)

№№	Group	Defense markers		
		Lysozyme, units/kg	Catalase, mkat/kg	API
1	Control	141±12	8,93±0,41	4,85±0,29
2	FSO	102±8 p<0,05	7,52±0,14 p<0,01	2,36±0,20 p<0,01
3	FSO + «Kverulin»	120±9 p>0,05; p_1>0,05	9,03±0,22 p>0,2; p_1<0,01	3,46±0,27 p<0,01; p_1<0,05
4	FSO + «Lysozyme-forte»	122±9 p>0,05; p_1>0,05	8,82±0,18 p>0,2; p_1<0,01	3,56±0,23 p<0,01; p_1<0,05
5	FSO + Dental elixir «Mint + amaranth extract»	127±10 p>0,1; p_1>0,05	9,10±0,16 p>0,2; p_1<0,01	4,08±0,30 p>0,05; p_1<0,01

p – compared to the group 1; p_1 – compared to the group 2.

The percentages of increase in the level of pathology markers and the percentages of decrease in the level of protection markers in the gums of rats (Fig. 1, 2) indicate a positive effect of herbal preparations on the condition of the gums.

Table 4 and fig. 4, 5 present the results of determining the level of biochemical markers of pathology and protection in periodontal bone tissue. These data show that in rats treated with FSO, the activity of AP significantly increases in bone tissue, but the activity of ALP, the MI index and the calcium content decrease. In rats that received herbal remedies on the background of FSO, the activity of acid and alkaline phosphatases in periodontal bone tissues, as well as calcium content, normalized. As for the MI index, all herbal remedies significantly increased it, and in rats that received applications with dental elixir, it was almost to the control level.

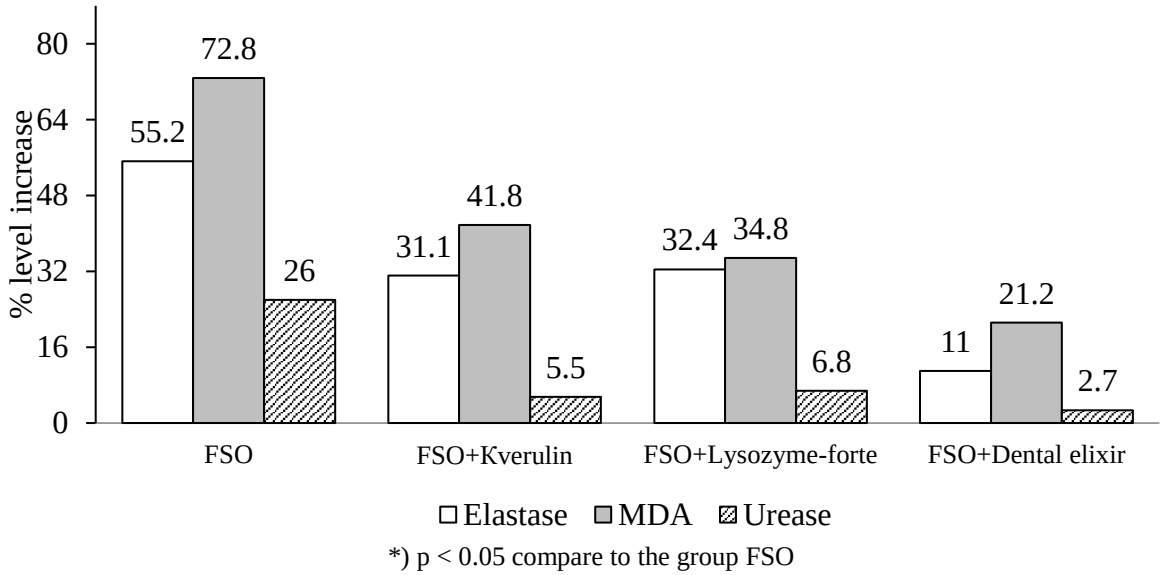

Fig. 1. The effect of herbal medicines on increasing the level of pathogenicity markers in the gums of rats fed fried sunflower oil (FSO)

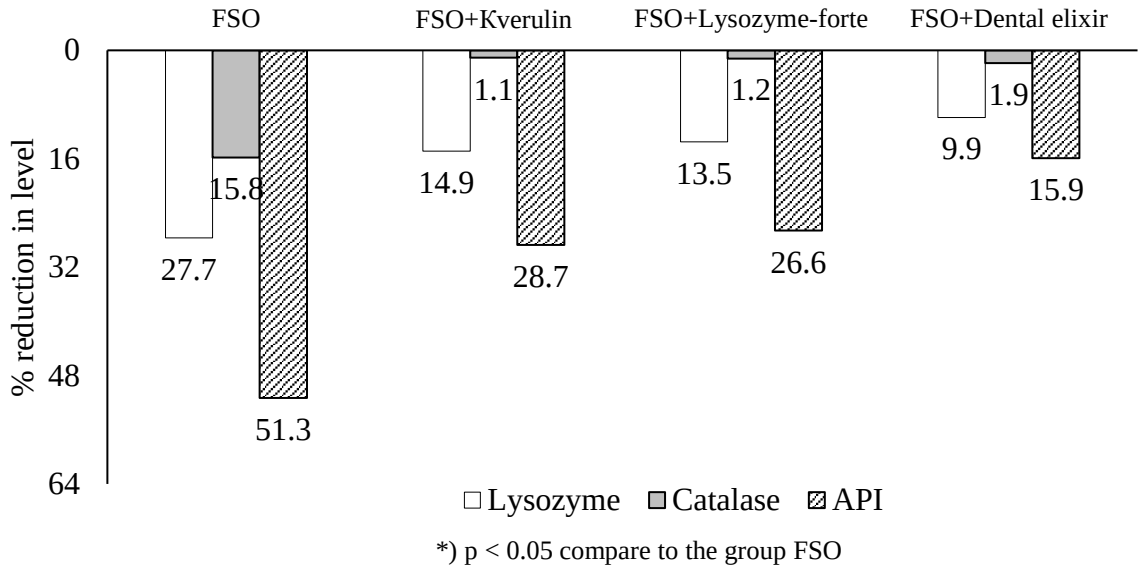

Fig. 2. The effect of herbal medicines on the reduction of protective markers in the gums of rats fed fried sunflower oil (FSO)

The total values of the level of pathology in the gums and in the periodontal bone tissue are presented in fig. 3, 6. From these data it is clear that the herbal preparation «Kvertuline» reduces the level of pathology markers in the gums by 2 times, and in the bone tissue — by 3 times. The herbal preparation «Lysozyme-forte» reduces the level of pathology in the gums by

2.3 times, and in the bone tissue – by 5 times. The dental elixir reduces the degree of pathology markers in the gums by 4 times, and in the bone tissue by almost 6.5 times.

Table 4. The effect of herbal preparations on biochemical markers of periodontal bone pathology in rats that consumed fried sunflower oil (FSO)

№№	Groups	Biochemical markers			
		Acid phosphatase (AP), μκ-κατ/κg	Alkaline phosphatase (ALP), μκ-cat/kg	Mineralization index (MI)	Calcium, mol/kg
1	Control	1,87±0,30	48,10±2,63	25,70±2,8	1,97±0,08
2	FSO	3,70±0,29 p<0,01	36,73±3,31 p<0,01	9,9±2,1 p<0,01	1,57±0,15 p<0,05
3	FSO + «Kverulin»	2,83±0,35 p>0,05; p_1<0,05	44,25±3,62 p>0,05; p_1>0,05	15,6±1,9 p<0,01; p_1<0,05	1,78±0,15 p>0,05; p_1>0,05
4	FSO + «Lysozyme-forte»	2,32±0,19 p>0,05; p_1<0,01	43,85±4,42 p>0,05; p_1>0,05	18,9±1,7 p<0,01; p_1<0,05	1,93±0,17 p>0,1; p_1>0,05
5	FSO + Dental elixir «Mint + amaranth extract»	2,30±0,21 p>0,1; p_1<0,05	47,05±4,42 p>0,1; p_1<0,05	20,4±2,0 p>0,05; p_1<0,01	1,91±0,13 p>0,1; p_1>0,05

p – compared to the group 1; p_1 – compared to the group 2.

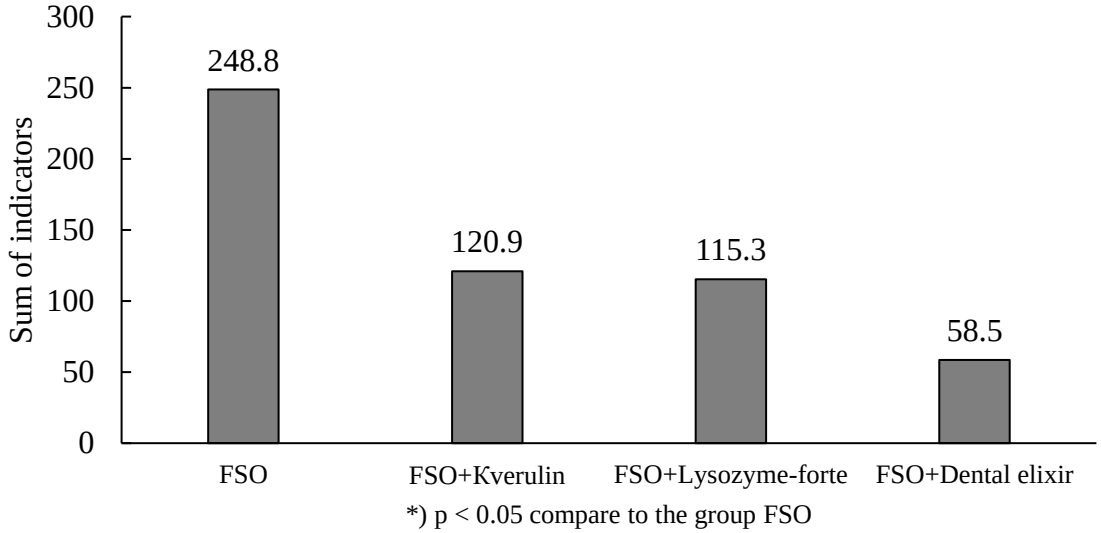

Fig. 3. The sum of indicators of the influence of herbal medicines on the level of pathogenicity and protection markers in the gums of rats fed fried sunflower oil (FSO)

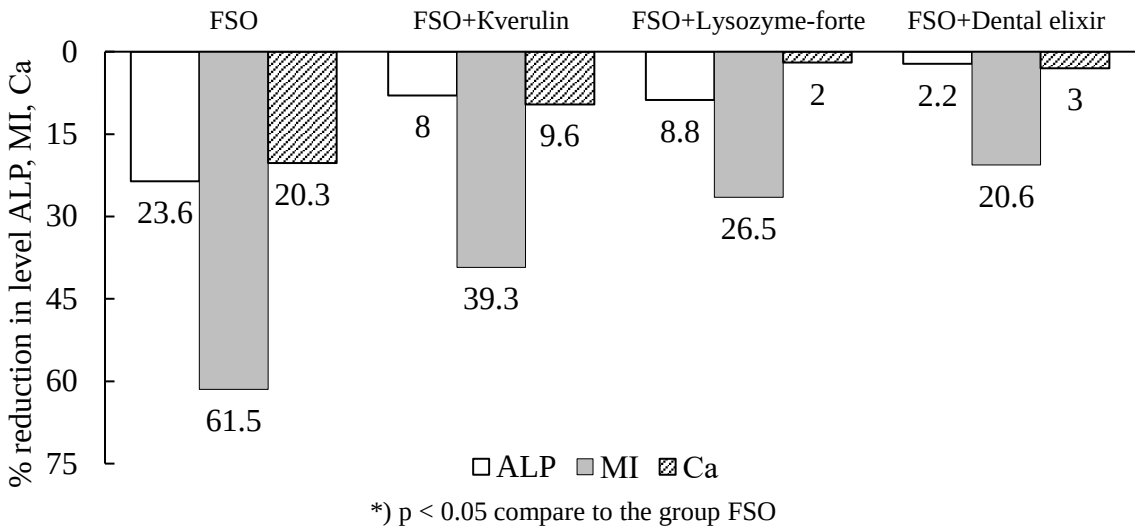

Fig. 4. The effect of herbal medicines on reduction in the level of biochemical markers (ALP, MI, Ca) in the periodontal bone tissue of rats that consumed fried sunflower oil (FSO)

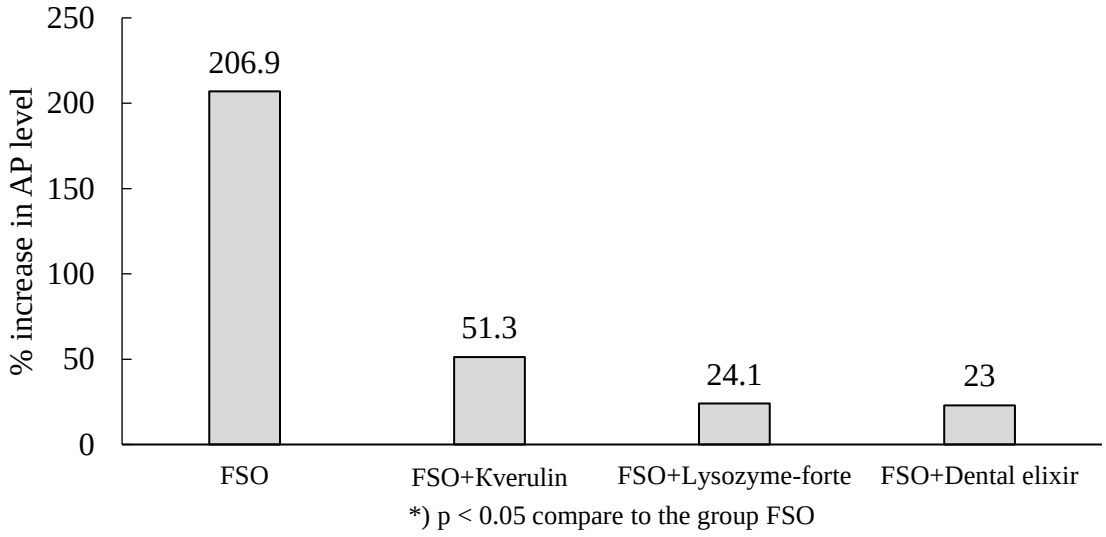

Fig. 5. The effect of herbal preparations on increasing the level of acid phosphatase in the periodontal bone tissue of rats that consumed fried sunflower oil (FSO)

The therapeutic and prophylactic effectiveness of herbal medicines calculated by us is presented in the fig. 7. From these data it is clear that herbal medicines have the most effective effect on periodontal bone tissue, and the most effective drug was the dental elixir «Mint + amaranth extract».

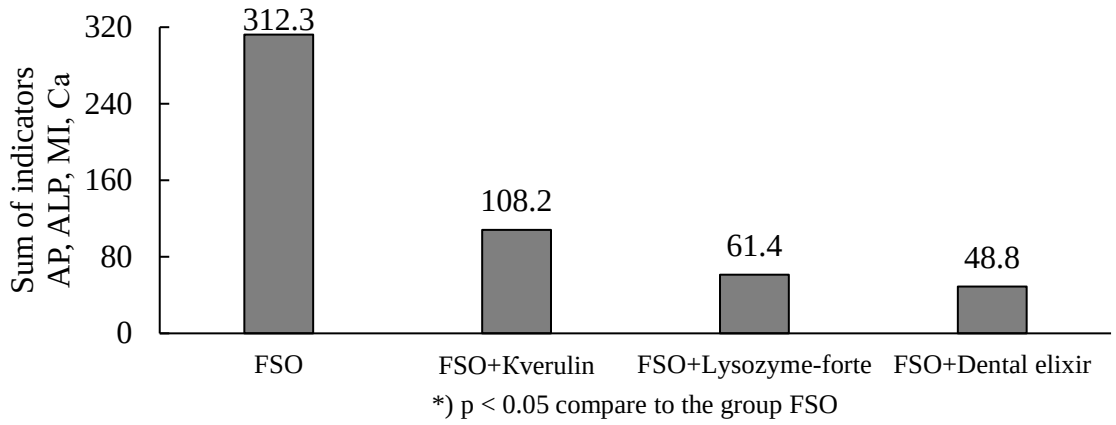

Fig. 6. The sum of indicators of the influence of herbal medicines on the level of biochemical markers in the bone tissue of the periodontium of rats fed fried sunflower oil (FSO)

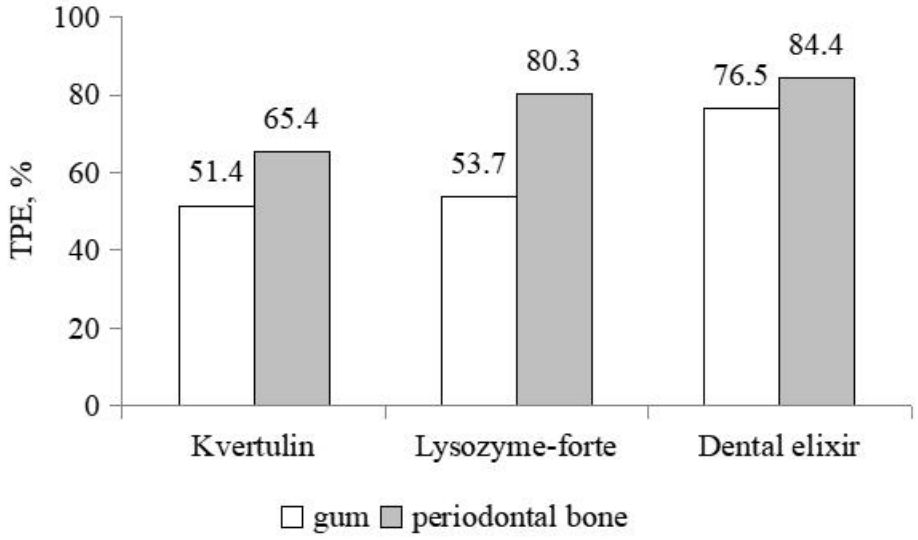

Fig. 7. Therapeutic and prophylactic efficacy (TPE) of herbal preparations in periodontal disease in rats receiving fried sunflower oil

The total pathology index of the gums of rats that consumed FSO was 248.8 %, while in the liver of rats it was almost 700 % [13]. Biochemical markers of pathology in both tissues were the same, so such a large difference in the total pathology index may indicate a certain resistance of the soft tissues (mucous membrane) of the periodontium to the action of FSO.

We also draw attention to the fact that periodontal bone tissue, even with a smaller number of biochemical markers (only 4), turned out to be more sensitive to the pathogenic effect of FSO: the total pathology index was 312.3 %.

At the same time, the gums were more sensitive to the therapeutic and prophylactic effects of herbal medicines than the liver [13]. Thus, «Kvertuline» in the liver of rats receiving FSO reduced the total pathology index by 20 %, while in the gums it reduces the total pathology index by 51 %. The situation is similar with other herbal medicines

The most effective therapeutic and prophylactic herbal preparation was the dental elixir «Mint + Amaranth extract», which includes mint extract, amaranth seed extract, bioflavonoid hesperidin, prebiotic inulin, calcium citrate, detergent cetavlon. These experimental data make it possible to recommend the dental elixir «Mint + Amaranth extract» for clinical use for the prevention of periodontitis.

Conclusion

1. Consumption of fried sunflower oil (FSO) by rats causes an increase in the activity of elastase and MDA content in the gums of rats and tendency to increase urease activity and a decrease in the level of lysozyme, catalase, and the antioxidant-prooxidant index API.

2. Consumption of FSO by rats leads to an increase in acid phosphatase activity in periodontal bone tissue with a simultaneous decrease in alkaline phosphatase activity, calcium content, and a significant decrease in bone mineralization activity.

3. The use of herbal remedies «Kvertuline», «Lysozyme-forte» and dental elixir «Mint + amaranth extract» against the background of FSO reduces the level of pathology markers in the gums (elastase, MDA) and normalizes the level of protection markers (lysozyme and catalase).

4. In periodontal bone tissue, the use of the above herbal remedies normalizes the level of acid and alkaline phosphatases and calcium content, however, only the dental elixir normalizes the MI index.

5. To assess the therapeutic and prophylactic efficacy (TPE) of herbal medicines, a comprehensive biochemical method was used, based on taking into account the nature of changes in biochemical markers of pathology and protectionin periodontal tissues.

6. The most effective in terms of TPE was the dental elixir «Mint + Amaranth extract», followed by the dietary supplements «Lysozyme-Forte» and «Kvertuline».

Author Contributions

The authors agree on equal distribution of partial participation.

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Informed Consent Statement

Informed consent was obtained from all subjects involved in the study.

Data Availability Statement

All information is publicly available and data regarding this particular patient can be obtained upon request from corresponding senior author.

Conflicts of Interest

The authors declare no conflict of interest.

Acknowledgments

The authors declare that there are no conflicts of interest.

References

1. Bouchard P, Carra MC, Boillot A [and others]. Risk factors in periodontology: a conceptual framework. Journal of Clinical Periodontology. 2017;44(2): 125-131.

2. Bartold PM, Van Dyke TE. Host modulation: controlling the inflammation to control the infection. Periodontol. 2017;75(1):317-329. doi: 10.1111/prd.12169.

3. Levitsky AP, Reizvikh OE. The correlation of dysbiosis and stomatological diseases in children (review). East European Scientific Journal (Wschodnioeuropejskie Crasopismo Nankowe). 2016;1(5(9):99-103. (in Russian).

4. Levitsky AP. Digestive enzymes of the salivary glands: thesis abstract... Dr. Biol. Sci.. – Odessa, 1974:53. (in Russian).

5. Gereljuk VI, Nejko NV, Pavljuk TD. Osteotropic therapy in periodontology. Guidelines. – Ivano-Frankivs'k, 2000:23. (in Ukrainian)

6. Pogrebnjak GV. Changes in the alveolar bone of female rats after ovariectomy: experimental correction of detected disorders. Ukrainian dental almanac. 2003;1:48-55. (in Ukrainian).

7. Levitsky AP. Fried butter: benefit or harm? – Odesa: Ecology, 2021:42. (in Russian).

8. Marcellin P, Kutala BK. Liver diseases: A major, neglected global public health problem requiring urgent actions and large-scale screening. Liver Int. 2018;1:382-386. doi: 10.1111/liv.13682

9. Levitsky AP, Egorov BV, Lapinskaya AP [and others]. Inadequate fat diet. Journal of Education, Health and Sport. 2020;10(7):248-255. DOI http://dx.doi.org/10.12775/JEHS.2020.10.07.029

10. Levitsky AP, Potapova IL. Fatty food, fatty acids, Healthy sunflower olive. Intern. Journ. Food a Nutrition. Sciences. 2015;4(3):15-20.

11. Levitsky AP. Disbiotic syndrome: etiology, pathogenesis, clinic, prevention and treatment. Dentistry Bulletin. 2019;10:14-20. (in Russian).

12. Levitsky AP, Makarenko OA, Denga OV [and others]. The experimental methods of the study of osteogenesis stimulators. Guidelines. – Kiev, 2005:50. (in Russian).

13. Levitsky A, Selivanska I, Velychko V. Integrated biochemical determination of the therapeutic and preventive effectiveness of herbal remedies for liver damage in rats that consumed fried sunflower oil. Fitoterapiia. Chasopys – Phytotherapy. Journal. 2024;3:86-91 doi: https://doi.org/10.32782/2522-9680-2024-3-86