Levitsky A. P., Pupin T. I., Selivanskaya I. O., Lapinskaya A. P., Markov A. V. Reduction of mineralizing activity of periodontal bone tissue in rats with consumption of ordinary sunflower oil. Journal of Education, Health and Sport. 2022;12(12):323-330. eISSN 2391-8306. DOI http://dx.doi.org/10.12775/JEHS.2022.12.12.049
https://apcz.umk.pl/JEHS/article/view/41499
https://zenodo.org/record/7470097

The journal has had 40 points in Ministry of Education and Science of Poland parametric evaluation. Annex to the announcement of the Minister of Education and Science of December 1, 2021. No. 32343. Has a Journal's Unique Identifier: 201159. Scientific disciplines assigned: Physical Culture Sciences (Field of Medical sciences and health sciences); Health Sciences (Field of Medical Sciences and Health Sciences).

Punkty Ministerialne z 2019 - aktualny rok 40 punktów. Załącznik do komunikatu Ministra Edukacji i Nauki z dnia 1 grudnia 2021 r. Lp. 32343. Posiada Unikatowy Identyfikator Czasopisma: 201159. Przypisane dyscypliny naukowe:Nauki o kulturze fizycznej (Dziedzina nauk medycznych i nauk o zdrowiu); Nauki o zdrowiu (Dziedzina nauk medycznych i nauk o zdrowiu).

Received: 22.11.2022. Revised: 05.12.2022. Accepted: .21.12.2022.

UDC 616.71+616.003+579.26

REDUCTION OF MINERALIZING ACTIVITY OF PERIODONTAL BONE TISSUE IN RATS WITH CONSUMPTION OF ORDINARY SUNFLOWER OIL

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Abstract

More than 20 % of people over the age of 50 suffer from osteoporosis. Lipids play an important role in the pathogenesis of osteoporosis.

Background. To investigate the effect of ordinary (high-linoleic) sunflower oil (OSO) on the state of periodontal bone tissue.

Methods. In 5 series of experiments, determine the mineralization activity of the alveolar appendix of the mandible rats that were obtained with feed 5 or 15 percent of OSO for from 22 to 75 days. Mineralizing activity was determined by the ratio of activity of alkaline and acid phosphatase. In liver lipids (fractions of phospholipids and free fatty acids) were determined by the content of long-chain polyunsaturated fatty acids (LCPUFA) by the gas chromatographic method.

Results. A decrease in mineralizing activity in OSO consumption, especially when using a feed with 15 % of OSO against a dysbiosis or metabolic syndrome. A significant

(almost 10 times) is shown to reduce the content of ω-3 PUFA in fraction of phospholipids of the liver of rats receiving a diet with a content of OSO.

Conclusion. Sunflower oil reduces the mineralizing activity of periodontal bone tissue by reducing the endogenous biosynthesis of ω-3 PUFA.

Keywords: osteoporosis; bone mineralizing activity; periodontal disease; sunflower oil; ω-3 LCPUFA.

Introduction

Ordinary sunflower oil (OSO) forms the basis of the fat diet of the population of Ukraine. The high palatability of this oil and the economic possibilities of obtaining it in large quantities from sunflower seeds, which is the most common oil crop in Ukraine, led to its wide consumption in home cooking and in the food industry for the production of various fat-containing products.

Unfortunately, OSO contains a large amount of linoleic acid (from 50 to 65 %), which at low levels exhibits a number of negative properties.

First, the presence of two double bonds ($C_{18:2}$, ω-6) in the linoleic acid molecule determines its instability during storage and thermal cooking due to peroxidation processes [1, 2].

Secondly, linoleic acid is inferior to oleic acid (the main acid of olive oil) in the rate of metabolism in mitochondria, where the energy of the fatty acid is converted into the energy of ATP [3].

Thirdly, linoleic acid inhibits endogenous biosynthesis of ω-3 polyunsaturated fatty acids (PUFA), which occurs at the expense of endogenous microbiota [4, 5].

Fourth, in our experiments it was shown that the consumption of OSO significantly increases the degree of periodontal bone tissue atrophy and tooth decay [6, 7].

And finally, fifthly, linoleic acid in the body is transformed into arachidonic acid ($C_{20:4}$, ω-6), from which pro-inflammatory eicosanoids are formed [1, 3].

It is very likely that the pathogenic effect of OSO is due to its ability to reduce the endogenous biosynthesis of ω-3 PUFAs, which have a positive effect on the state of bone tissue not only of the periodontium, but also of the bones of the spine and limbs [8, 9].

The purpose of our study was to determine the effect of OSO on the state of periodontal bone tissue by such an indicator as mineralizing activity. Bone mineralizing activity is easily determined by the ratio of the activity of alkaline phosphatase (ALP), which

is a biochemical marker of osteoblasts and osteocytes, and acid phosphatase (AP), which is a biochemical marker of osteoclasts [10, 11].

Materials and research methods

Food refined sunflower oil was used in the work, the fatty acid composition of which is presented in Table 1. Biological experiments were conducted on white Wistar rats in five series of experiments (Tables 2 and 3). After euthanasia of the animals under thiopental anesthesia, the lower jaw was isolated and in the homogenate of the alveolar process, the activity of ALP and AP was determined by the hydrolysis of p-nitrophenyl sodium phosphate at pH 10.5 and pH 4.8, respectively [12].

Table 1. Fatty acid composition of ordinary (high-linolenic) sunflower oil

Fatty acid	Short formula	Contents, %
Lauric acid	$C_{12:0}$	0
Myristic acid	$C_{14:0}$	0,15
Palmitic acid	$C_{16:0}$	9,74
Stearic acid	$C_{18:0}$	3,90
Oleic acid	$C_{18:1}$ ω-9	30,6
Linoleic acid	$C_{18:2}$ ω-6	53,5
Linolenic acid	$C_{18:3}$ ω-3	0,03
Arachinic acid	$C_{20:0}$	0,20
Eicosenoic acid	$C_{20:1}$	0,22
Arachidonic acid	$C_{20:4}$ ω-6	0
Eicosapentaenoic acid	$C_{20:5}$ ω-3	0
Docosapentaenoic acid	$C_{22:5}$ ω-3	0
Docosahexaenoic acid	$C_{22:6}$ ω-3	0

The content of ω-3 LCPUFA (by amount $C_{20:4}$ ω-3 + $C_{22:5}$ ω-3 and $C_{22:6}$ ω-3) in the fractions of phospholipids and free fatty acids (FFA) was determined in the liver homogenate by the gas chromatographic method [13, 14].

In experimental series No. 3, thermoperoxide sunflower oil (TPSO) was used [2]. In the experimental series No. 4, feed with 15 % OSO was used against the background of experimental dysbiosis, which was caused by the antibiotic lincomycin [15]. In experimental

series No. 5, feed with 15 % OSO was used against the background of experimental metabolic syndrome (+ lincomycin + cytostatic) [15].

Table 2. Composition of rations for rats [12]

Components	Free fat diet (FFD)	Fatty diet (FD)
Corn starch	65	60(50)
Defatted soybean meal	20	20
Ovalbumin	6	6
Saccharose	4	4
Mineral mixture	4	4
Vitamin mixture	1	1
Ordinary sunflower oil (OSO)	0	5(15)

Table 3. Characterization of experimental series on feeding white rats with ordinary sunflower oil (OSO) FFD – fat-free diet

Series of experiments	Number of rats	Age of rats, months.	The sex of rats	% OSO in feed	Associated pathology	Duration of feeding, days
1. Free fat diet						
1.1. FFD	7	5-6	males	0	absent	30
1.2. Diet with OSO	7	5-6	males	5	absent	30
2. Free fat diet						
2.1. FFD	6	6-7	males	0	absent	75
2.2. Diet with OSO	6	6-7	males	6	absent	75
3. Free fat diet						
3.1. FFD	6	6-7	males	0	absent	75
3.2. Diet with thermoperoxide OSO	7	6-7	males	6	peroxide intoxication	75
4. High-fat diet						
4.1.FFD	7	3	males	0	absent	22
4.2. Diet with OSO	8	3	males	15	dysbiosis	22
5. High-fat diet						
5.1. FFD	7	4-5	males	0	absent	40
5.2. Diet with OSO	7	4-5	males	15	metabolic syndrome	40

Results and discussion

Table 4 presents the results of determining the activity of ALP, AP and the mineralizing index (ALP/AP) in the periodontal bone tissue of rats treated with OSO. From the presented data, it can be seen that in all cases of using feed containing OSO, the activity of alkaline phosphatase significantly decreases and the activity of acid phosphatase increases significantly, which indicates a significant decrease (by 2-4 times) in the mineralizing activity of periodontal bone tissue.

Table 4. Phosphatase activity and mineralizing index in periodontal bone tissue of rats that consumed OSO

Series of experiments	Alkaline phosphatase (ALP) μ-cat/kg	Acid phosphatase (AP), μ-cat/kg	Mineralizing index MI=ALP/AP
1.1. FFD	128,9±7,3	3,75±0,33	34,4±2,8
1.2. FD (5 % OSO)	93,2±4,1 p<0,05	5,80±0,49 p<0,05	16,1±1,7 p<0,01
2.1. FFD	130,5±6,4	4,03±0,45	32,4±3,3
2.2. FD (6 % OSO)	82,6±7,0 p<0,05	6,71±0,83 p<0,05	12,3±1,2 p<0,01
3.1. FFD	115,4±6,8	3,11±0,40	37,1±4,0
3.2. FD (4 % TPSO)	36,7±3,3 p<0,01	3,70±0,19 p>0,05	9,9±0,9 p<0,01
4.1. FFD	110,3±6,1	3,10±0,52	35,6±3,1
4.2. FS (15 % OSO)	33,9±4,0 p<0,01	9,22±0,39 p<0,01	8,9±0,9 p<0,01
5.1. FFD	119,6±7,2	3,24±0,54	36,9±4,0
5.2. FD (15 % OSO)	67,3±12,6 p<0,05	5,85±0,97 p<0,05	11,5±1,2 p<0,01

Table 5 presents the results of determining the content of long-chain PUFA (LCPUFA) in the fractions of phospholipids and free fatty acids of liver lipids of rats that received food with OSO.

Table 5. The effect of consumption of OSO dietary on the content of LCPUFA in phospholipids and FFA of rat liver

Experimental series	Σ ω-6 PUFA, %	Σ ω-3 PUFA, %	ω-6/ω-3 PUFA
Phospholipids			
1.1. FFD	11,28	2,33	4,84
1.2. FD (5 % 3CO)	11,96	0,23	52,00
FFA			
1.1. FFD	17,71	3,90	4,54
1.2. FD (5 % OSO)	14,52	1,49	9,74

As can be seen from these data, the largest amount of LCPUFA is in the fraction of FFA, and ω-6 LCPUFA, represented by arachidonic acid, predominate in terms of quantity.

The consumption of feed with a content of 5 % OSO of dietary has little effect on the content of ω-6 LCPUFA, but significantly reduces the content of ω-3 LCPUFA, represented by eicosapentaenoic, docosapentaenoic and docosahexaenoic acids. Thus, the content of ω-3 LCPUFA in the FFA fraction decreases by 2.6 times, and in the phospholipid fraction by 10 times. As a result, the ratio of ω-6/ω-3 LCPUFA in the fraction of FFA increases by more than 2 times, and in the fraction of phospholipids - by almost 11 times.

Analyzing the data we received about the negative impact of the consumption of ordinary sunflower oil on the state of periodontal bone tissue, we have certain reasons to consider the following mechanism of their influence quite likely.

Firstly, linoleic acid, which is the basis of OSO, can inhibit the growth of probiotic bacteria (bifidumbacter and lactobacter) [4, 5]. Secondly, there is every reason to consider probiotic bacteria as producers of endogenous ω-3 LCPUFA.

Thirdly, it is known that ω-3 LCPUFA have a positive effect on the bone mineral density of both the periodontium and the skeleton [16]. It has been established that ω-3 LCPUFA activate the membrane enzyme Ca^{2+}+-ATPase, which ensures calcium transport.

The negative impact of OSO on the state of bone tissue that we have established can be considered as one of the main reasons for the increase in the incidence of periodontitis in Ukraine.

Suppression of endogenous biosynthesis of ω-3 LCPUFA with consumption of dietary fiber may be one of the reasons for the significant increase in cardiovascular and neuro-endocrine diseases in the population of Ukraine.

The data we received provide grounds for recommendations to reduce the consumption of ordinary sunflower oil and replace it with fats with a low content of linoleic acid.

Conclusion

Consumption of ordinary (high-linoleic) sunflower oil reduces the mineralizing activity of periodontal bone tissue due to a significant decrease in the endogenous biosynthesis of ω-3 LCPUFA.

Given that the decrease in the mineral density of the periodontal bones is one of the reasons for the development of periodontitis, we consider it expedient to reduce the consumption of OSO for the purpose of prevention.

Acknowledgments

The authors declare that there are no conflicts of interest.

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