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INFLUENCE OF HIGH-FAT NUTRITION WITH DIFFERENT FAT-ACID COMPOSITION OF FATS ON LIPID PEROXIDATION PROCESSES IN RAT'S ORGANS AND TISSUES

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Abstract

The aim. To determine the effect of high-fat diet (HFD) using fats of different fatty acid composition on the content of malondialdehyde (MDA) in the organs and tissues of rats and on catalase activity.

Materials and methods. The following edible fats were used: regular (high linoleic) sunflower oil, high oleic sunflower oil, palm, butter and coconut oils at a concentration of 15 % by weight of the diet. The rats were fed for 64 days. Before euthanasia, rats were bled from *v. porta* and *v. cava inferior*. The MDA content and catalase activity were determined in blood serum, in liver homogenates, intestinal mucous membranes, in skeletal muscles, heart and brain. The antioxidant-prooxidant index of API was calculated from the ratio of catalase activity and MDA content.

Results. found that the content of the MDA in *v. cava* is significantly higher than *v. porta*. High-fat diet (HFD) increases the MDA content in all tissues except the brain, and most of all in the liver and after the consumption of high-palmitic fats (palm and butter), as well as high-linoleic sunflower oil. The exception is high oleic sunflower oil, the consumption of which does not increase the MDA content. Catalase activity reacts little to HFD.

Conclusions. the liver secrete MDA into the blood. HFD increases the level of MDA in organs and tissues, except for the brain, without significantly reducing the activity of catalase. The antioxidant effect of HFD using high oleic sunflower oil can be explained by the antioxidant properties of oleic acid.

Keywords: high-fat diet, lipid peroxidation, malondialdehyde, catalase, high oleic sunflower oil

Introduction

High-fat diet (HFD) has become extremely common in recent decades [1-4], and to some extent changed the composition of dietary fats due to the increasing use of palm oil [5-8] and new vegetable oils with altered fatty acid composition, obtained by selection methods [9]. Consumption of animal fats, which contain a significant amount of saturated fatty acids, in particular palmitic [10], has also increased significantly.

It has been established that HFD can cause the development of dysbiosis and systemic inflammation in the body [11, 12] and this pathogenic effect of dietary fats largely depends on their fatty acid composition [10, 13].

Peroxidation of unsaturated fatty acids can be involved in the mechanism of pathogenic action of HFD, which produces various toxic substances, one of the end products of which is malonic dialdehyde (MDA) [14-16].

Objective of this work was to study the content of MDA in the organs and tissues of rats that have long received high-fat diets (HFD) with dietary fats that have different fatty acid composition. The state of the antioxidant system was also determined, the marker of which was the activity of the catalase enzyme and the API index (antioxidant-prooxidant index as the ratio of catalase activity and MDA content).

Methods

The following dietary fats were used in the work:

– Unrefined sunflower oil, frozen, pressed (manufacturer firm "Smak solnca" "Marchenko V.V.", Ukraine);

High-oleic sunflower oil "Olivka", TS U 15.4 13903778-36:2002 (manufacturer of SPA "Odessa Biotechnology", Ukraine) [17];

Peasant butter 72.5 % fat (manufacturer PCF "Agromarin", Ukraine);

– Palm oil (manufacturer "Dukees RBD", Malaysia);

– Coconut oil of the Bees brand (producer of PGFO Edible Oils SDN BHD, Malaysia).

The fatty acid composition of these fats was determined by gas chromatographic method [18]. The results of determining the content of fatty acids are presented in table 1. These data show that the

main fatty acid of ordinary sunflower oil is linoleic $(C_{18:2})$, high-oleic sunflower oil "Olivka" contains 88 % oleic acid $(C_{18:1})$, in butter and palm oil contains approximately equally palmitic $(C_{16:0})$ and oleic acids, and coconut oil contains the most lauric acid $(C_{12:0})$.

Feeding experiments were performed on white Wistar rats (males, 8-9 months, live weight 240-260 g), which were divided into 6 equal groups of 6 heads each. Group 1 received a standard balanced diet containing 5% feed fat (almost 90 % are linoleic, oleic and palmitic acids). Groups 2-a – 6-a received HFD, in which 15 % of the grain component was replaced by the appropriate amount of test fat: 2nd group – sunflower oil, 3rd – high-oleic sunflower oil, 4th – butter, 5th – palm oil and 6th – coconut oil. The duration of feeding was 64 days.

Before euthanasia of rats under thiopental anesthesia (20 mg / kg) received blood from v. porta and v. cava inferior, and then carried out total bleeding from the heart. The liver, mucous membranes of the small and large intestines were isolated, skeletal muscle samples were obtained, and the heart and brain were isolated.

In serum, in homogenates of tissues and organs, the content of MDA was determined by thiobarbitur method [20] and catalase activity [21]. According to the ratio of catalase activity and MDA content, the antioxidant-prooxidant index of API was calculated by the formula [19]:

$API = \frac{A_{kat} \cdot 10}{C_{MDA}}$

The results of the experiments were subjected to standard statistical processing [22].

Results

In fig. 1 presents the results of determining the increase in live weight of rats for 64 days of feeding HFD.

From these data it is seen that the consumption of ordinary sunflower oil, butter or palm oil caused an increase in live weight, respectively, in 2; 2.2 and 2.5 times. Consumption of coconut oil had little effect on weight gain and consumption of high-oleic sunflower oil had no effect at all.

Table 2 presents the results of determining the organ index of the liver in rats consumed HFD. Significantly increases the organ index of the liver of rats, which consumed of ordinary sunflower oil and

palm oil. The organ index of the liver also increases in rats that consumed butter (however, p>0.05). Only coconut oil and high-oleic sunflower oil did not affect the organ index of the liver.

Table 3 presents the results of determining the content of MDA in serum with *v. porta* and *v. cava* inferior rats consumed HFD. From these data it is seen that the content of MDA in the blood *v. porta* does not depend significantly on the nature of the fatty diet, whereas in the blood *v. cava* MDA content is 2-3 times higher than in the blood with *v. porta*, which may indicate the injection of MDA by the liver of rats. Moreover, the level of MDA increment by the liver does not depend significantly on the amount of dietary fat, but depends on the nature of the fat component: most when consuming palm oil, less consumption of ordinary sunflower oil and significantly lower when consuming high oleic sunflower and coconut oils.

In fig. 2 and 3 present the results of determining the effect of HFD on the content of MDA in the organs and tissues of rats. In control rats, which received a low-fat diet (only 5 % fat), the highest content of MDA was observed in the brain (65-70 mmol/kg), and the lowest – in the mucous membrane of the small and large intestine (5-6 mmol/kg). In the liver, skeletal muscle and heart, the MDA content was 25-35 mmol/kg. HFD in all organs and tissues increases the MDA content except the brain, in which all dietary fats tend to decrease the MDA content, especially butter and coconut oil.

The largest increase in the content of MDA in the liver of rats treated with HFD, especially with palm oil. Only HFD with high oleic sunflower oil does not increase the content of MDA in the liver, mucous membranes of the digestive tract, skeletal muscle and heart. HFD with palm oil increases the content of MDA not only in the liver, but also in the mucous membranes of the small and large intestines and skeletal muscle. The content of MDA in skeletal muscle significantly increases HFD with the use of ordinary sunflower oil.

In the heart, only HFD with butter significantly increases the MDA content

Table 4 presents the results of determining the activity of the antioxidant enzyme catalase. The greatest activity of this enzyme is observed in the liver and heart, and the least – in the mucous membrane of the colon. In the conditions of HFD,

this indicator shows only a tendency to decrease its level (significantly only in the mucous membrane of the small intestine). It can be assumed that the level of catalase does not have a significant effect on the level of MDA. It is possible that other antioxidant systems in the body are more relevant to the state of lipid peroxidation.

A more sensitive indicator of the state of antioxidant systems is the API index, the results of which are presented in Fig. 4 and 5.

As can be seen from these data, all dietary fats (except high oleic sunflower oil) reduce the API index. In the brain, do not reduce the API index of any of the dietary fats.

Thus, our studies have shown that HFD activates the processes of lipid peroxidation (LP), as evidenced by a significant increase in the content of the final product of LP – malonic dialdehyde. The most sensitive organ was the liver, in which all fats, except high-oleic sunflower oil, increased the level of MDA. In other tissues, an increase in MDA levels was observed only with HFD involving regular sunflower oil and palm oil (skeletal muscle) or butter (heart).

Only in the brain did HFD cause a decrease in the MDA content, despite the fact that the brains of healthy rats contain significantly more MDA than all other tissues.

The increase in MDA content in HFD leads to a significant decrease in the API index, although the activity of the antioxidant enzyme catalase is slightly reduced after HFD. It is possible that other antioxidant systems of the body play a decisive role in counteracting LP. This can be the glutathione system, the selenium system, tocopherol, and many other systems.

In any case, based on the obtained data, we can say that to prevent the activation of LP, it is necessary to use high-oleic sunflower oil in the diet [17], because oleic acid performs an antioxidant function in the body [23].

Conclusions

1. The liver is an organ that produces and secretes malonic dialdehyde.

2. HFD stimulates LP activity in most organs and tissues (especially in the liver). In the brain, HFD reduces the MDA content.

3. The most effective way to increase the content of MDA is the consumption of HFD with the content of high palmitic fats (palm oil and butter), as well as high linoleum sunflower oil.

4. High oleic sunflower oil does not increase the MDA content, possibly due to the antioxidant properties of oleic acid.

Acknowledgments

The authors declare that there are no conflicts of interest.

References

- Sharafetdinov, Kh.Kh., Plotnikova, O.A. Obesity as

 Global Challenge of the 21st Century: Nutritional Care, Prevention and Therapy. Nutrition issues. 2020; vol. 89; № 4; 161-171 (in Russian)
- Ivashkin, V.T., Maevskaya, M.V. Lipotoxicity and metabolic disorders in obesity. Russian Journal of Gastroenterology, Gepatology, Coloproctology. 2010; vol. 20, № 1; 4-13 (in Russian)
- Romancova, T.I. The obesity epidemic: truthful and veritable reasurs. Obesity and metabolism.
 2011; № 1; 1-14 (in Russian)
- Gozhenko, A.I., Gryshko, Ju.M. Pathogenetic basis of the obesity development as a consequence of functional-metabolic imbalance in the organism (review). Actual problems of transport medicine. 2019; № 1(55); 29-40 (in Ukrainian)
- 5. Titov, V.N., Rozhkova, T.A., Amelyushkina, V.A. (et al.). The role of almitic fatty acid in the initiation of hypertriglyceridemia, hypercholesterolemia, atherosclerosis and atheromatosis. International Medical Journal. 2015; vol. 21; № 2 (82); 5-14 (in Russian)
- 6. Titov, V.N. Excess palmitic fatty acid in food the main cause of lipoidosis of insulindependent cells: skeletal myocytes, cardiomyocytes, periportal hepatocytes, Kupffer macrophages and β-cells of the pancreas. Clinical laboratory diagnostics. 2016; vol. 61; № 2; 68-77 (in Russian)
- Wang, X., Jiang, X., Deng, B. (et al.). Lipopolysaccharide and palmitic acid synergistically induced MCP-1 production via MAPK-meditated TLR4 signaling pathway in

RAW264.7 cells. Lipids Health Dis. 2019; vol. 18; 71.

- 8. Gozhenko, A. I., Levitsky, A. P., Stepan, V. T., Pustovoit, I. P., Badiuk, N. S., Maslyukov, A. K. Advantages of high olein sunflower oil over palm oil according to biochemical research results. PhOL – PharmacologyOnLine; N 2; P. 293-301; https://pharmacologyonline.silae.it/files/archives /2020/vol2/PhOL_2020_2_A028_Gozhenko.pdf
- 9. Lisitsyn, A.N. Scientific support of modem technologies for the production of new types of oil and fat products. Oilseeds. 2012; № 2; 151-152 (in Russian)
- Levitsky, A.P. Pathophysiology of high-fat nutrition and ways to prevent its complications. Bulletin of XVII readings. V.V. Podvysotsky, May 24-25, 2018, Odessa; 120-124 (in Russian)
- Velichko, V.I., Tkachuk, V.V., Levitsky, A.P. Development of dysbiosis in tissues of rats fed with a high fat food. J. Health sciences. 2014; vol. 4, № 12; 84-92 (in Russian)
- Levitsky, A.P., Khodakov, I.V., Levchenko, E.M. Influence of high fat diets with different composition of fatty acids on the content of essential fatty acids in liver lipids. Journal of Education, Health and Sport. 2015; vol. 5, №12; 598-607 (in Russian)
- 13. González-Beceira, K., Ramos-Lopez, O., Barrón-Cabrera, E. (et al.). Fatty acids, epigenetic mechanisms and chronic diseases: a systematic review. Lipids Health Dis. 2019; vol. 18; 178.
- 14. Voskresenskiy, O.N., Levitsky, A.P. Peroxide lipids in the living organism. Questions of medical chemistry 1970; vol. 16, № 6; 563-583 (in Russian)
- 15. Khajuria, A. Lipid peroxidation. Everyman's Sci. 1997; vol. 32, № 3; 109-113.
- 16. Sharapov, M.G., Novoselov, V.I. The Catalytic and signaling-regulatory role of peroxiredoxins in carcinogenesis. Overview. Biochemistry. 2019; vol. 84, № 2; 147-171 (in Russian)
- 17. Levitsky, A.P. Olivka: the unique sunflower oil, the analogue to olive oil. Odessa: KP OGT; 2013; 28 (in Russian)
- 18. Levitsky, A.P., Makarenko, O.A., Khodakov, I.V. Methods to investigate fats and oils. Odessa: KP OGT; 2015; 32 (in Russian)
- 19. Levitsky, A.P., Makarenko, O.A., Demyanenko, S.A. Methods of experimental dentistry

- (teaching aid). Simferopol: Tarpan, 2018; 78 (in Russian)
- 20. Stalnaya, I.D., Garishvili, T.G. The method of revelation of malonic dialdehyde with thiobarbituric acid. Moskva: Meditsina, 1977; 66-68 (in Russian)
- 21. Girin, S.V. The modification of the method of the determination of catalase activity in biological substrates. Laboratory diagnosis. 1999; Nº 4; 45-46 (in Russian)
- 22. Trukhacheva, N.V. Mathematical statistics in medical and biological research using Statistica packages. M.: GEOTAR-Media, 2012; 379 (in Russian)
- 23. Titov, V.N., Lisitsyn, D.M. Regulation of peroxidation in vivo as a stage of inflammation. Oleic acid, reactive oxygen species invaders and antioxidants. Clinical laboratory diagnosis. 2005; № 6; 3-12 (in Russian)

Fatty acid	Abbreviated formula	Sunflower oil	High oleic sunflower oil	Butter	Palm oil	Coconut oil
Caprylic	C _{8:0}	0	0	1,25	0	2,00
Capric	C _{10:0}	0	0	2,67	0	3,02
Lauric	C _{12:0}	0	0	2,97	0,19	46,57
Myristic	C _{14:0}	0,15	0,03	10,43	1,16	22,70
Palmitic	C _{16:0}	9,74	4,44	<u>27,88</u>	<u>42,02</u>	11,67
Stearic	C _{18:0}	3,90	3,07	12,73	4,87	13,60
Oleic	C _{18:1}	30,60	<u>88,06</u>	<u>26,61</u>	<u>40,93</u>	0,30
Linoleic	C _{18:2}	<u>53,46</u>	1,21	3,08	9,49	0,02
Linolenic	C _{18:3}	0,03	0,11	0,53	0,17	0
Arachinic	C _{20:3}	0,20	0,27	0,28	0,47	0,12
Eicosenic	C _{20:1}	0,22	0,16	0,12	0,16	0
Arachidonic	C _{20:4}	0	0	0,05	0	0
Behenic	C _{22:0}	0,72	1,07	0	0,13	0
Lignoceric	C _{24:0}	0,25	0,81	0	0,10	0

Table 1. Fatty acid composition of used dietary fats (% of the amount of fatty acids)

Table 2. The effect of HFD on the organ index of the liver of rats

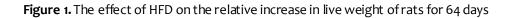
NºNº	Edible fat	Organ index of the liver, g/kg
1	Control	31,7±2,9
2	Sunflower oil	39,3±2,3 p<0,05
3	High oleic sunflower oil	35,9±4,1 p>0,3
4	Butter	38,7±3,2 p>0,05
5	Palm oil	42,8±3,1 p<0,05
6	Coconut oil	32,7±2,8 p>0,5

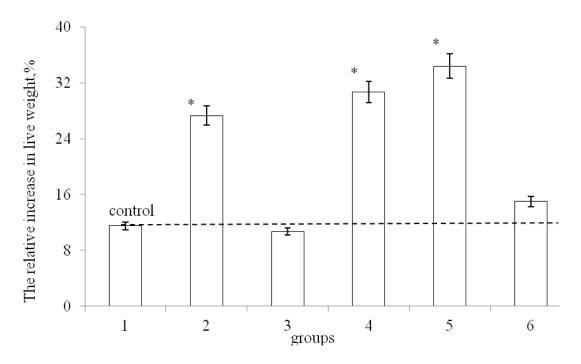
 Table 3. Secretion of MDA by the liver of rats treated with various dietary fats (15%)

NºNº	Croup	MDA, r	ΔMDA		
IN-IN-	Group	v. porta	v. porta	+	
1	Control	0,75±0,08	1,84±0,02	1,09±0,06	
2	Sunflower oil	0,78±0,04 p>0,3	2,15±0,07 p<0,01	1,37±0,05 p<0,05	
3	High oleic sunflower oil	0,27±0,08 p>0,05	1,82±0,05 p>0,3	0,85±0,04 p<0,01	
4	Butter	0,81±0,08 p>0,05	1,97±0,12 p>0,1	1,16±0,07 p>0,3	
5	Palm oil	0,88±0,04 p>0,05	2,51±0,10 p<0,01	1,63±0,09 p<0,01	
6	Coconut oil	0,78±0,06 p>0,3	1,55±0,07 p<0,05	0,77±0,06 p<0,01	

	Catalase activity, mcat/kg					
Nº Nº	Liver	Small intestine	Colon	Skeletal muscles	Heart	Brain
1 – Control	6,3±0,1	3,6±0,2	1,9±0,1	2,5±0,1	5,0±0,1	3,3±0,1
2 – Sunflower oil	6,1±0,1	2,2±0,3	1,6±0,1	2,2±0,1	4,7±0,1	3,0±0,1
	p>0,05	p<0,01	p>0,05	p>0,05	p>0,05	p>0,05
3 – High oleic sunflower oil	6,1±0,1	3,0±0,2	1,7±0,1	2,4±0,2	5,0±0,1	3,2±0,1
	p>0,05	p>0,05	p>0,05	p>0,3	p=1	p>0,3
4 – Butter	5,9±0,1	2,6±0,1	1,6±0,1	2,3±0,1	5,0±0,1	3,0±0,1
	p<0,05	p<0,01	p>0,05	p>0,05	p=1	p>0,05
5 – Palm oil	5,9±0,1	2,7±0,2	1,6±0,1	2,4±0,1	5,0±0,1	2,9±0,1
	p<0,05	p<0,01	p>0,05	p>0,3	p=1	p<0,05
6 – Coconut oil	6,1±0,1	2,9±0,2	1,6±0,1	2,3±0,1	4,8±0,1	2,9±0,1
	p>0,05	p<0,05	p>0,05	p>0,05	p>0,1	p<0,05

Table 4. The effect of HFD on catalase a	activity in the organs and tissues of rats
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1 - control, 2 - sunflower oil, 3 - high oleic sunflower oil, 4 - butter, 5 - palm oil, 6 - coconut oil

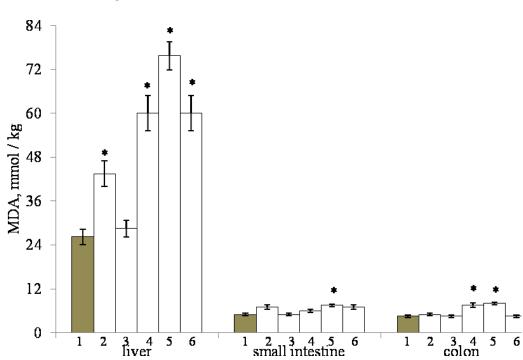


Figure 2. The effect of HFD on the level of MDA in rat tissues

1 - control, 2 - sunflower oil, 3 - high oleic sunflower oil, 4 - butter, 5 - palm oil, 6 - coconut oil

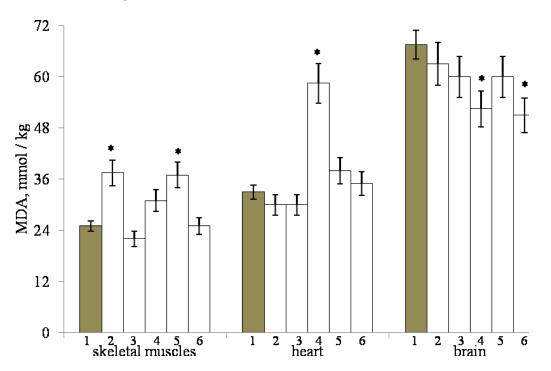
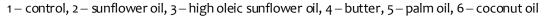
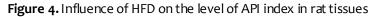
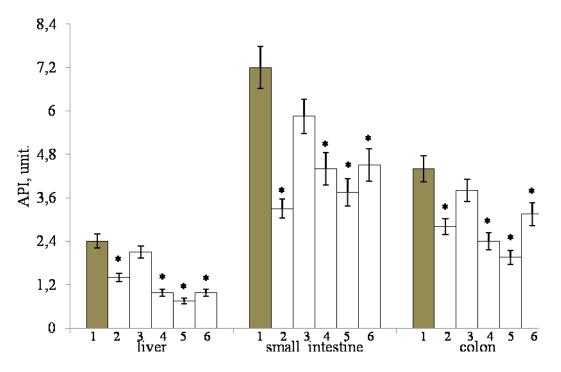


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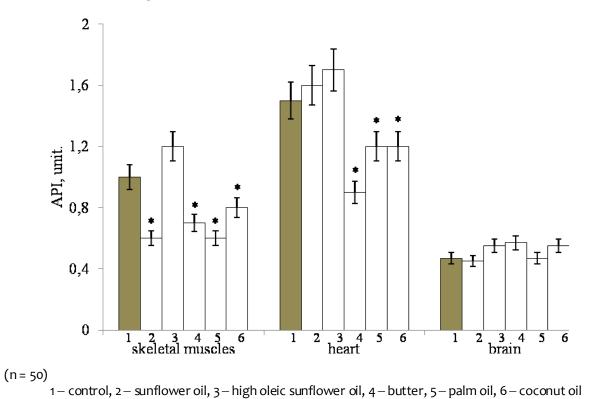


Figure 5. Influence of HFD on the level of API index in rat tissues