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LIPID PEROXIDATION AND ANTIOXIDANT PROTECTION IN PLASMA AND LIVER OF RABBITS

Дослідженнями пероксидаційних процесів було встановлено наявність підвищеного вмісту загальних ліпідів і ТБК-активних продуктів у плазмі крові та тканинах печінки кролів у 1- та 15-добовому віці, що засвідчило про активний перебіг процесів пероксидного окиснення ліпідів. Активність супероксиддисмутази, каталази, глутатіонпероксидази, вмісту відновленого глутатіону і церулоплазміну у плазмі крові та тканинах печінки у кролів 1- та 15-добового віку була незначною.

Дослідженнями активності та вмісту ензимів антиоксидантної системи у кролів 60-добового віку було зафіксовано підвищення активності каталази, глутатіонпероксидази, вмісту відновленого глутатіону і церулоплазміну у плазмі крові та тканинах печінки.

Ключові слова: кролі, пероксидне окиснення ліпідів, плазма крові, печінка, загальні ліпіди, ТБК-активні продукти, супероксиддисмутаза, каталаза, глутатіонпероксидаза, відновлений глутатіон, церулоплазмін.

Statement of the problem, analysis of recent research and publications. One of the important mechanisms for body normal development is to maintain lipid peroxidation balance and antioxidant protection state (AOP) [10, 11].

Under physiological conditions, the level of lipid peroxidation (LPO) is supported by anti- and pro-oxidante equilibrium system. LPO positive impact on the living body (structure restoring and maintaining biological membranes properties, participating in energy processes) is provided by antioxidant protection system, i.e. a set of enzymatic and non-enzymatic factors that protect cells from free radicals influence [10, 13, 14].

Speed and regulation of lipid peroxidation is being carried by multicomponent antioxidant system that provides binding and modifying of free radicals and prevents the formation and destruction of peroxides. Correlation of free radical oxidation intensity and antioxidant activity determines the antioxidant status of cells, tissues and body as a whole [10, 11].

A number of factors, primarily qualitative and quantitative composition of diet, largely determines the state of LPO and AOP processes [l, 3].

An important condition for increasing vitality and resistance of rabbit organism in modern industrial conditions of the field management is to maintain the physiological state of the body at different stages of individual development [1, 6].

Currently, the rabbit breeding has no efficient ways for antioxidant deficiency correction according to animal age. Therefore, the research on biochemical characteristics of lipid metabolism and antioxidant system activity in different age rabbits is urgent [1, 3].

The study of antioxidant balance and this process regulation changes in rabbits with individual development, makes interest due to the search for ways to influence growth, development, body functional state, fodder nutrients assimilation, productivity and quality of the end product.

The aim of our research was to study the lipid peroxidation and antioxidant protection parameters in blood plasma and liver tissues of rabbits, depending on age.

Material and methods. The experiment was carried out on rabbits of New Zealand breed, fed with full diet mixed fodder.

The after slaughter liver and blood from heart were taken for studies from of 1, 15, 30, 45, 60, 75, 90-days old rabbits. The content of total lipids [4], reduced glutathione [2], ceruloplasmin [12], TBA-active products [8], superoxide dismutase (SOD) [9], catalase [5] and glutathione peroxidase (GPO) [7] was determined in blood plasma and liver tissue homogenate.

The obtained numerical data was statistically processed by Microsoft EXCEL To determine the likely differences between the average values we used the Student test.

Results and discussion. Our findings indicate that the total lipids content in the rabbits blood plasma, from the first day of birth tended to increase and by 45-th day was significantly higher by

54.1 % as compared to 1-day old animals (Table 1). In further studies the rabbits (60-, 75-, and 90-days old) showed its slight fluctuation.

Table 1 – **Content of total lipids and TBA-active products in rabbits blood plasma and liver** (M ± m, n = 5)

Age, days	Total lipids		TBA-active products	
	plasma, g/dm^3	liver mg/g tissues	plasma, micromole/dm^3	liver, micromole/g tissues
1	21,79±3,71	26,83±0,77	4,82±0,25	0,648±0,02
15	23,10±1,04	25,30±0,90	6,55±0,44**	0,446±0,07*^
30	23,72±1,72	27,64±1,03	6,63±0,23	0,194±0,04*^^^
45	33,58±3,17**^	28,25±2,18	6,85±0,64	0,176±0,02^^^
60	16,23±3,02**	33,82±2,71^	5,85±0,38	0,193±0,03^^^
75	17,01±2,49	27,97±1,25	6,39±0,17	0,132±0,02^^^
90	19,47±1,93	27,81±1,47	6,77±0,59	0,117±0,01^^^

Note: here and further the difference is valid when * -p<0,05; ** -p<0,01;
*** – p<0,001 – as compared to previous age; ^ – p<0,05; ^^- p<0,01;
^^^ – p<0,001 – as compared to 1- day old animals.

In the liver of one day old rabbits there was found that the content of total lipids had a slight increase in animals after birth (Table 1). Later, a tendency was recorded to increase the content of total lipids in 30- and 45-days old rabbits. The maximum value of total lipids in the liver tissues of 60 days old rabbits was significantly higher by 26 % as compared to one day old animals. The 75- and 90-days old animals showed a downward tendency of the total lipids content in liver tissue.

By determining the content of TBA-active products in the blood plasma, there was also found a definite tendency to increase by this indicator, particularly in 1- to 45-days old rabbits (Table 1). There was also observed a credible difference of TBA-active products content in the blood plasma of rabbits by the 15th day, which was higher by 36% as compared with the one-day old animals.

A slight content increase of TBA-active products (Table 1) was noted in liver tissues of one day old rabbits. Further, the 15- and 30-days old animals showed credible decline of this indicator, as compared to the previous age. The content of TBA-active products in 15-days old rabbits was credibly lower by 1.5 times and in 30-days old ones -by 3.3 times as compared to one day old rabbits. Later, in liver tissues of 45-, 60-, 75- and 90-days old rabbits, the credible reduction of TBA-active products content continued. The content of TBA-active products credibly decreased in 45 days old rabbits by 3.7 times, in the 60 – by 3.4 times, the 75 – by 4.9 times and the 90 – by 5.5 times, as compared to one day old rabbits.

There were found changes of superoxide dismutase activity, which is one of the main animal antioxidants and protects the body cell membrane from the damaging effects of free radicals [1, 3]. In particular, in 15 days old rabbits blood plasma there was observed an increased activity of SOD, which was credibly higher by 53.8 % as compared to one day old rabbits (Table 2).

Table 2 – **Activity of antioxidant enzymes in blood plasma and liver of rabbits** (M ± m, n = 5)

Age, days	SOD		Catalase		GPO	
	plasma, unit/sm^3	liver, unit/g tissue	plasma, mcat/sm^3	liver, cat/g tissue	plasma, micromole×min/dm^3	liver, micromole× xb/g tissue
1	25,11±2,84	1,83±0,47	303,70±12,70	44,29±0,11	1,83±0,01	31,41±0,40
15	38,63±4,62 *^	1,30±0,62	318,88±10,08	45,67±0,72	1,86±0,01	33,46±0,28 **^^
30	51,71±5,48 ^^	3,50±0,86	374,29*10,53 **^^	46,30±0,39 ^^^	1,88±0,03	32,44±0,29 *^
45	67,35±8,23 ^^	6,23±0,27 ^^^	575,96±6,34 ***^^^	42,44±0,90 **	1,92±0,02 ^	30,79±0,42 **^^
60	98,36±5,10 **^^^	3,84±1,48	588,48±9,71 ^^^	43,36±0,64	1,94±0,03 ^^	31,18±0,29
75	53,87±5,00 ***^^^	4,49±1,06 ^^	609,79±10,87 ^^^	40,99±0,29 **^^^	1,79±0,01 **	31,61±0,52
90	59,30±6,31 ^^^	4,87±0,76 ^^	489,38±13,82 ***^^^	43,45±0,53 **	1,69±0,04 *^^	33,00±0,41 *^

The SOD activity increase was observed in 60 days old rabbits and during this period amounted 98,36 ± 5,10 units / cm3, which was credibly higher by 1.5 times as compared to the previous age (45 days) and by 3,9 times – as compared with one day old animals.

The superoxide dismutase activity in liver tissues had small increase in one day old rabbits (Table 2). In 30 days old rabbits liver there was observed a tendency to SOD activity increase, and in 45 days old animals this indicator had high value and was higher by 3,4 times ($p<0,001$) as compared to one day old animals. The SOD activity decrease tendency was detected in liver tissues of 60 days old rabbits.

SOD is a key antioxidant protection enzyme, it restores superoxide radical to a less toxic hydrogen peroxide, protects cell membranes from the negative effects of free radicals. Since SOD utilizes active oxygen forms to make H_2O_2, it is important for cell functioning to establish a balance between the SOD activity and catalase [6].

Catalase plays an important role in redox reactions, therefore its activity increase in the blood of 75 days old rabbits (Table 2) is the evidence of active peroxidation processes in the young body [3, 6]. The catalase activity on the 30-th and the 45-th day had a credible difference and were higher as compared to previous periods animals and as compared to one day old animals by 23.2 % and by 89.6 % respectively.

The liver tissues catalas activity in the study tended to increase in 1, 15 and 30 days old animals. In particular, the 30 days old rabbits catalase activity was credibly higher by 4.5 % as compared with the one day old animals, in subsequent studies (45-, 60-, 75-, 90-days old) a certain tendency to decrease the enzyme activity was observed.

The glutathione peroxidase activity level, which owns an active role in protecting the lysosomal cell membranes from peroxidation, also had a tendency to increase in blood plasma during 1-, 15-, 30-, 45- and 60-days of rabbits life (Table 2). The highest activity was recorded in the 60 days age. Activation of the enzyme in the blood of animals is only possible on condition of maintaining a sufficiently high level of intracellular glutathione (GSH), which serves not only as the reaction substrate, but also as factor, necessary for the permanent restoration of selenole groups, placed in the catalytic center of the enzyme, which is oxidized in the glutathione peroxidase reactions [1, 3, 6, 11].

Changes of glutathione activity in the liver tissues were characterized by credible increase of this enzyme activity by 6.5 % in 15-days old rabbits and by 3.3 % in 30 days old rabbits compared to one day old animals. During 45-, 60-, 75- and 90 days of life the GPO values were characterized by a tendency to gradual growing and balanced antioxidant enzyme activity.

The content of restored glutathione in blood plasma of rabbits during the experiment was characterized by small fluctuations (Table 3).

The study of restored glutathione content in the liver showed credible increase in 15-days old rabbits by 1.5 times, as compared with one day old animals. Further, the restored glutathione content in 60 days old rabbits was by 2.3 times higher in comparison with 45-days old animals and by 1.9 times, compared to one day old rabbits.

Table 3 – **Restored glutathione and ceruloplasmin content in blood plasma and liver of rabbits** (M ± m, n = 5)

Age, days	GSH		Ceruloplasmin	
	plasma, micromole/dm^3	liver, micromole/g tissues	plasma, mg/dm^3	liver, mg/g tissue
1	0,27±0,01	0,26±0,03	107,10±5,89	0,91±0,14
15	0,28±0,01	0,38±0,03*^	134,75±10,40*^^	0,96±0,11
30	0,22±0,03	0,34±0,02	22U20±12,30***^^^	1,89±0,17***^^
45	0,19±0,02^^	0,22±0,03**	523,43±27,65***^^^	2,01±0,15^^^
60	0,20±0,03^	0,50±0,06**^	533,23±17,19^^^	2,62±0,15*^^^
75	0,15±0,01^^^	0,34±0,07	494,73±l 8,65^^^	2,54±0,22^^^
90	0,24±0,03*	0,32±0,02	456,93±10,92^^^	1,74±0,14**^^

There was reported a ceruloplasmin content credible increase in the blood plasma of rabbits from 1- to 60 days age (Table 3). Maximal high amount of ceruloplasmin was found in 60 days old rabbits, which was credibly higher, as compared to one day old rabbits. These changes may indicate the increased metabolic processes, where ceruloplasmin playsan important role, enhancing the rabbits body antioxidant defense [1, 3, 11, 14].

Ceruloplasmin content in the liver of 1- and 15-days old rabbits tended to increase. In subsequent experiment periods (30, 45 and 60-days) this trend continued. In particular the ceruloplasmin content in 30-days old rabbits was significantly higher by 2 times, as compared with the previous period and by 2.1 times, as compared with one day old animals. The highest content of ceruloplasmin was detected in the liver of 60-days old rabbits, which was credibly higher by 2,9 times, as compared with one day old animals.

The determined changes of lipid peroxidation and antioxidant defense system indicators certify the tension of prooxidant-antioxidant balancing of young rabbits. In particular, this phenomenon can be explained by the age-related characteristics of AOC formation in young rabbits body [1, 3, 6, 10, 14].

Conclusions and recommendations for further research. In order to maintain rabbits herd and improve their performance, it is necessary to control the content of total lipids, TBA-active products and activity of antioxidant enzymes (superoxide dismutase, catalase, glutathione peroxidase, glutathione and ceruloplasmin) in animal blood plasma and liver tissues.

Incresed level of total lipids and TBA-active products in blood plasma and liver tissue in 1- and 15-days old rabbits testified to the active peroxidation processes that may be a consequence of the young body stress response to environmental factors in adaptation period.

The 60 days old rabbits had increase of catalase activity, glutathione peroxidase, restored glutathione content and ceruloplasmin in their blood plasma and liver tissue, reflecting the acquisition of young body antioxidant resistance.

The further research and study of lipid peroxidation and antioxidant protection depending on the age dynamics of different breeds rabbits is concidered topical.

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Перекисное окисление липидов и антиоксидантная защита в плазме крови и печени кроликов

М.Н. Федорченко

Исследованиями пероксидационных процессов было установлено наличие повышенного содержания общих липидов и ТБК-активных продуктов в плазме крови и тканях печени кроликов в 1- и 15-суточном возрасте, что свидетельствует об активном ходе процессов пероксидного окисления липидов. Активность супероксиддисмутазы, каталазы, глутатионпероксидазы, содержания восстановленного глутатиона и церулоплазмина в плазме крови и тканях печени у кроликов 1- и 15-суточного возраста была незначительной.

Исследованиями активности и содержания энзимов антиоксидантной системы у кроликов 60-суточного возраста было зафиксировано повышение активности каталазы, глутатионпероксидазы, содержания восстановленного глутатиона и церулоплазмина в плазме крови и тканях печени.

Ключевые слова: кролики, пероксидное окисление липидов, плазма крови, печень, общие липиды, ТБК-активные продукты, супероксиддисмутаза, каталаза, глутатионпероксидаза, восстановленный глутатион, церулоплазмин.

Lipid peroxidation and antioxidant protection in plasma and liver of rabbits

M. Fedorchenko

The research on peroxidation processes showed the presence of higher concentrations of total lipids and TBA-active products in blood plasma and liver tissue in 1- and 15-days old rabbits, which testified for active processes of lipid peroxidation. The superoxide dismutase, catalase, glutathione peroxidase, glutathione content and ceruloplasmin activity in plasma and liver tissue of 1- and 15-days old rabbits was negligible.

The research on activity and enzymes content of antioxidant system in 60-days old rabbits showed increased activity of catalase, glutathione peroxidase, restored glutathione content and ceruloplasmin in blood plasma and liver tissue.

Key words: rabbits, lipid peroxidation, blood plasma, liver, total lipids, TBA-active products, superoxidedismutase, catalase, glutathione peroxidase, restored glutathione, ceruloplasmin.

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