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Nutritional Status and Indicators of Oxidative Stress among End-Stage Renal Disease Patients Treated with Continuous Ambulatory Peritoneal Dialysis

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Abstract

Background: The objective of the study was to determine the oxidative stress (OS) intensity depending on the nutritional status (NS) among end-stage renal disease patients treated with continuous ambulatory peritoneal dialysis (CAPD) and to investigate the effectiveness of medical strategies for the correction of nutritional disorders (ND).

Methods: 69 end-stage renal disease patients treated with CAPD were examined who had varying degrees of ND. General clinical, biochemical parameters, OS markers were identified. Basing on the obtained data, the level of OS markers was determined in groups of patients with different NS. Subsequently, patients with moderate and severe ND were randomly assigned to two groups. The first group (n=20) included patients who received in complex treatment additionally to traditional treatment of CAPD Levocarnitine and one exchange per day of intraperitoneal fluid with amino acids. The second group consisted of patients (n=20) who received instead of one Dianeal fluid intraperitoneal fluid with amino acids.

Results: OS indicators were increased in all four groups of patients with different NS, but they were the highest among patients with moderate and severe malnutrition. After the treatment the patients of the first study group had a statistically significant decrease in the MDA content, both in blood serum and erythrocytes ($p<0.005$). At the same time, the analysis of the informative markers dynamics for antioxidant oxidative stress (AOS) of blood serum allowed to register a statistically credible increase in their mean values among patients after treatment ($p<0.05$). It should be emphasized that no statistically significant effect of Levocarnitine on the anthropometric parameters of nutritional status and serum albumin level was obtained. However, after the therapy in the study group the values of SGA and protein consumption with food increased ($p<0.05$). At the same time, the patients from second study group had no positive effect on the reduction of oxidative stress, except for the level of transferrin ($p<0.05$) and contributes to the increase of serum albumin level ($p<0.05$).

Keywords: Chronic renal disease; Renal replacement therapy; Peritoneal dialysis; Nutritional status; Oxidative stress

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Introduction

One of the links in the development of chronic kidney disease (CKD), is a violation in the balance between oxidative and antioxidant (O/A) reactions toward the excess formation of lipid peroxides, the number of which correlates with the severity of the disease. Excessive activation of free radical processes with subsequent damage to the membrane structures of the body systems cells is one of the causes of high morbidity, decreased life quality and mortality in the specified population of patients [1,2].

Although peritoneal dialysis (PD) is a more biocompatible modality of dialysis compared to hemodialysis (HD), among patients treated with PD, oxidative stress intensity is likely to be higher compared with both the general population and patients with end-stage renal disease pre-dialysis stages [3-5]. In addition, the imbalance of O/A reactions has a significant effect on the state of chronic inflammation and, consequently, the subsequent development of peritoneal fibrosis among patients treated with PD [6,7]. The fluid composition used in the treatment of PD (low pH, increased osmolality, increased concentration of lactate and glucose breakdown products) affects the accumulation of oxidative products [8,9]. Huh et al. in their study, have suggested that the accumulation of glucose degradation molecules in glucose-containing peritoneal fluids can cause the onset and development of OS by enhancing the activity of nitric oxide synthase [10]. This hyperexpression of NOS among PD patients receiving treatment for more than 18 months has also been associated with increased peritoneal membrane calcification, increased vascular endothelial growth factor activity, and accumulation of major advanced glycation endproducts (AGE) [11,12].

Terawaki H, et al. (2007) in their study have demonstrated that oxidized albumin, another marker of OS status, was significantly increased among 21 PD patients compared with the healthy control group. The authors concluded that serum albumin oxidation may be

facilitated by small and medium uremic toxins among patients treated with CAPD [13-15].

Many studies have shown that OS increase among PD patients occurs on the background of PD-associated peritonitis, loss of residual renal function (RRF), nutritional disorders (ND) [16-18].

Xiao C, et al. (2018)having examined 64 patients treated with peritoneal dialysis method, have proved that serum albumin level, a surrogate marker of nutritional disorders (ND), directly correlates with OS, it means patients with hypoalbuminemia have significantly higher OS levels indicators than patients with normal blood serum albumin content [19]. Other studies have confirmed their assumptions and stated that OS correction among patients with protein-energy malnutrition (PEM) contributes to the improvement of nutritional status (NS), namely the increase of serum albumin level, subjective global assessment (SGA), body mass index (BMI) and lean body mass [20-22].

Therefore, considering the available research results, it can be affirmed that chronic inflammation and endothelial damage resulting from OS amplification are important precursors of nutritional disorders among patients with chronic renal disease.

It is important to have objective criteria for correcting O/A disorders to choose the optimal treatment regimen for these patients concerning inhibition of OS intensity. Nowadays, one of such therapeutic strategies is the use of medicines containing Levocarnitine. Levocarnitine is involved in most energy processes, its presence is obligatory for the oxidation of fatty acids, amino acids, carbohydrates, and ketone bodies, it stimulates the metabolism of components in the β-oxide conversion of free fatty acids in hepatocytes mitochondria, biosynthetic processes, normalizes indicators of lipid peroxidation (LP). These medicinal drugs are also used to increase muscle mass, thereby improving nutritional status indicators [23].

At the same time, a fluid enriched with amino acids was developed for patients treated with PD. Its main action is the correction of PEM. However, considering its composition, this fluid can also be considered as a method of OS correction, although there are few studies that would aim to study this effect [24].

In view of the abovesaid, it is necessary and appropriate to carry out studies aimed at studying the intensity of OS processes depending on the NS, and the effectiveness of therapeutic strategies for the ND correction.

Objective

To determine the intensity of OS depending on the nutritional status of end-stage renal disease patients treated with peritoneal dialysis. To investigate the effectiveness of therapeutic strategies for correcting nutritional disorders.

Material and Methods

The observational prospective open randomized study included 69 end-stage renal disease patients treated with peritoneal dialysis during 2012-2018 at the Kyiv Scientific and Practical Center of Nephrology and Dialysis which is the clinical base of the State Institution "Institute of Nephrology of National Academy of Medical Sciences of Ukraine".

The study was conducted in three stages. At the first stage of the study, all patients fulfilled standard clinical diagnostic methods, which included general clinical biochemical and instrumental methods of NS examination and certain markers of OS. Among OS markers, the concentration of by products of lipid peroxidation (LP) - malonic dialdehyde (MDA) in the blood by reaction with thiobarbituric acid was investigated; the concentration of ceruloplasmin by reaction with paraphenylenediamine dihydrochloride and transferrin by reaction with ammonium citrate. Basing on the data obtained, the antioxidant blood capacity (ABC) and OS index (IOS) were calculated. Research blood was taken from the ulnar vein in the morning after an 8h fast. At the second stage of the study the patients were divided into 4 groups, depending on the nutritional status, which was previously determined using laboratory, functional and anthropometric methods. The first group (n=20) included patients without malnutrition, the second group (n=10) included patients with a light degree of malnutrition, the third (n=20) and the fourth group (n=19) included patients with a moderate and severe degree of nutritional disorders, respectively. A comparative analysis of OS indicators was performed, both in the study groups and in the control group. At the third stage of the study, patients with moderate and severe PEM degrees were randomly assigned into two groups. The first group (n=20) included patients who received in complex treatment additionally to traditional treatment of CAPD intraperitoneal fluid with amino acids and Levocarnitine. This medicinal drug was prescribed intravenously at a dose of 0.5 g per day for 10 days per month for 3 months. The intraperitoneal fluid with amino acids was used once a day instead of one Dianeal fluid. The second group consisted of patients (n=20) who received instead of one Dianeal fluid only intraperitoneal fluid with amino acids once a day for 3 months.

Statistics

All received data have been processed using modern methods of descriptive statistics by means of theStat Soft Releases STATISTICA Version 10 (TULSA, OK, USA). Average value (M) and standard deviation (SD) were determined. For the statistical analysis, we used Student's *t*-test and nonparametric (*U*-test) Mann-Whitney test. Pearson's correlation tests were used to evaluate the association between oxidative stress biomarkers and nutritional parameters. Chi-square tests were used for comparison of two groups. The difference was reliable when the significance level was $p<0.05$.

Results

The average age of patients was 52.7±13.1 years, male was 66.6% (46 patients). According to the type of kidney damage, patients with glomerulonephritis prevailed and comprised 37 people, diabetic nephropathy - 24, the rest of the nosology - 8 people. The proportion

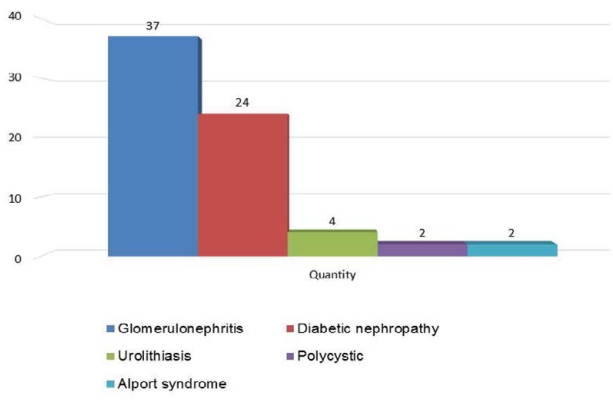

Figure 1: Proportion of causes of ESRD in the study population.

of patients by causes of ESRD is shown in the figure (Figure 1). In the process of OS markers determination, it was stated that in the examined population of end-stage renal disease patients treated with CAPD the concentration of OS markers in comparison with the conditionally healthy individuals is increased (Table 1).

Note: *indicates the statistically credible difference in comparison with the control group is $p<0.05$

The MDA content, namely, the metabolite of the prooxidant system increased 4.61 times ($p<0.001$) in blood serum and 1.01 (10.5%) times in erythrocytes compared with the reference group of conditionally healthy individuals, and ceruloplasmin 1,42 times. Along with an increase of the MDA production in the blood serum of end-stage renal disease patients, it was registered a decrease in the blood serum concentration of transferr in 2.38 times ($p<0.01$) and SH groups by 48% in blood serum and by 10.5% in erythrocytes ($p<0.05$) compared to similar indicators in the reference group. The IOS calculation showed an increase in OS intensity of almost 6.04 times in the blood serum and 1.23 times (23.5%) in erythrocytes.

Where: ESRD refers as End stage of renal disease

Conducting of the correlation analysis allowed to establish the presence of a reliable strong negative relationship between serum albumin level and MDA of blood serum (r=-0.8830; p=0.0000) (Figure 2).

Where: MD Arefers as Malonic dialdehyde and CAPD refers as Continuous ambulatory peritoneal dialysis.

Table 1: Oxidative stress parameters among end-stage renal disease patients treated with continuous ambulatory peritoneal dialysis.

Indicator		PD patients (n=69)	Conditionally healthy individuals (n=30)
Blood serum	MDA, mcM/l	569.6±188.8*	119±35*
	Transferrin, g/l	2.01±0.68*	5,0±1.0*
	Ceruloplasmin, g/l	0.29±0.09*	0.218±0.011*
	SH-groups, mcM/l	1.55±0.35*	2.22±0.02*
	ABC, rel.units	0.821±0.03*	1±0.085*
	IOS, rel.units	6.24±0.91*	1.036±0.04*
Erythrocytes	MDA, mcM/l	623.7±52,3*	549±51*
	SH-groups, mcM/l	19.4±1.05*	21.22±0.40*
	IOS, rel.units	1.28±0.04*	1.036±0.07*

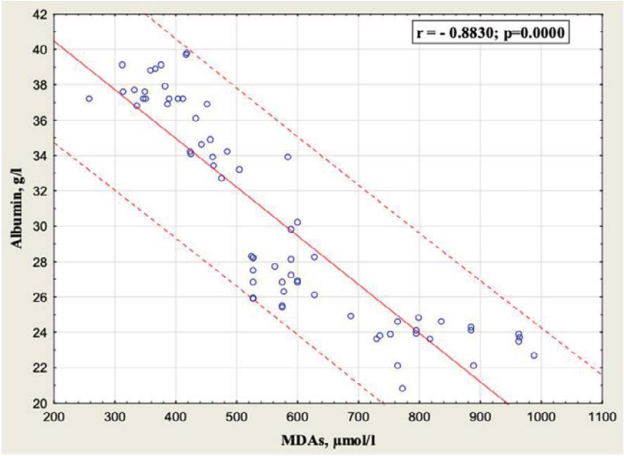

Figure 2: Linear correlation between serum albumin and serum MDA level in end-stage renal disease patients treated with CAPD.

For further analysis, the patients were divided into four groups according to the nutritional status determined based on laboratory, anthropometric and functional methods. Peculiarities of informative NS markers in groups are given in the table (Table 2).

The levels of OS indicators were subsequently determined depending on the degree of nutritional disorders (Table 3).

According to the obtained data OS indicators were found to be increased in all four groups but they were the highest among the patients with moderate and severe malnutrition. The ceruloplasmin concentration in blood serum while comparing the groups did not change significantly ($p>0.05$). However, the content of MDA serum among patients with severe malnutrition was 2.36 times higher than in the group of patients with normal NS. Along with the increase in the production of MDA, in the group of patients with severe nutritional disorders there was revealed a decrease in the concentration of blood serum for transferrin in 2.4 times, SH-groups in 1.62 (62%) and ABC in 0.61 (46.8%) times compared with similar indicators of the group of patients with normal NS. At the same time, as severe malnutrition was present, an increase of IOS was found 4 times more compared with the group with normal NS.

[1] P_1-P_2=0.0003; P_2-P_3=0.0001; P_3-P_4=0.027; P_1-P_4=0.00001

[2] P_1-P_2=0.003; P_2-P_3=0.001; P_3-P_4=0.0001; P_1-P_4=0.0001

[3] P_1-P_2=0.765; P_2-P_3=0.906; P_3-P_4=0.624; P_1-P_4=0.886

Table 2: Indicators of nutritional status in the studied population of patients.

Indicator	Sex	Groups of nutritional status state			
		No malnutrition (n=20)	Light degree (n=10)	Moderate degree (n=20)	Severe degree (n=19)
Body mass index, kg/m^2		25.0-19.0	22.4±3.32	20.8±3.04	19.7±2.9
Skin-fat fold over triceps at the point of circumference measurement, mm	Men	10.5-9.5	9.8±2.17	10.1±1.4	9.9±1.8
	Women	14.5 - 13	13.8±1.9	13.6±2.1	13.1±0.89
Shoulder muscles circumference, cm	Men	25.7-23	21.5±1.4*	19.1±0.8*	17.4±0.65*
	Women	23.5-21	21.3±1.22*	18.4±0.78*	16.3±0.63*
Albumin, g/l		>35	34.2±2.7*	29.7±2.12*	25.1±1.87*
The absolute number of lymphocytes, thousand in mql		>1.8	1.7±0.11*	1.51±0.07*	0.89±0.16*
Transferrin, g/l		>2.0	1.97±0.43*	1.8±0.21*	1.6±0.12*
SGA, points		7	4.42±1.28*	3.43±1.24*	1.29±0.47*
Proteins, g/kg/round the clock		>1.2	1.08±0.03*	0.89±0.04*	0.82±0.06*
Fats, g/kg/round the clock		1.1-1.3	1.26±0.07	1.24 ± 0.07	1.3 ± 0.03
Carbohydrates, g/kg/round the clock		4.2-4.5	4.24± 0.25	4.32±0.17	4.29±0.31
Dietary calories, kcal/kg/ day		30-35	32.8±1.9	32.7±1.04	32.5 ±1.68

Note: The statistically credible difference between groups - $p<0.05$*

Table 3: OS indicators among end-stage renal disease patients treated with CAPD depending on the indicators of nutritional status.

Indicator	Groups of nutritional status state			
	No malnutrition (n=20)	Light degree (n=10)	Moderate degree (n=20)	Severe degree (n=19)
MDAc[1], mcM/l	370.11±46.8	472.4±46.5	569.2±35.3	831.2±91.1
Transferrin[2], rel.units	2.97±0.79	1.94±0.07	1.73±0.07	1.32±0.2
Ceruloplasmin[3], g/l	0.27±0.04	0.31±13	0.3±0.09	0.289±0.09
SHc[4]-groups, mcM/l[4]	2.01±0.2	1.62±0.11	1.44±0.08	1.16±0.09
ABC[5], rel.units	0.94±0.025	0.85±0.037	0.82±0.056	0.64±0.017
IOS[6], rel.units	3.11±1.34	4.67±0.89	6.23±0.79	12.46±34

Note: Inter-group authenticity

[4]P_1-P_2=0.0003; P_2-P_3=0.002; P_3-P_4=0.0003; P_1-P_4=0.0001

[5]P_1-P_2=0.005; P_2-P_3=0.235; P_3-P_4=0.004; P_1-P_4=0.0001

[6]P_1-P_2=0.034; P_2-P_3= 0.005; P_3-P_4=0.005; P_1-P_4=0.0001

All these changes of O/A indicators influenced OS intensity, as a lack of antioxidant parameters formed a decrease in total antioxidant capacity of blood, contributed to the maintaining of high concentration of lipoperoxidation products (MDA) and their prolonged negative effects on the body which is evidenced by the high IOS indicators.

To correct nutritional disorders among end-stage renal disease patients treated with CAPD, an intravenous drug Levocarnitine and intraperitoneal fluid with amino acids were selected. Both groups were representative by the main demographic, social, clinical and laboratory indicators, the severity of nutritional disorders, and the duration of CAPD (Table 4).

Dynamics of NS parameters and OS markers among patients with moderate and severe levels of malnutrition in association with Levocarnitine usage and intraperitoneal fluid with amino acids on the parameters of NS and OS among patients with moderate and severe malnutrition are presented in the table (Table 5).

The data of the table demonstrate clearly the positive influence of the above complex therapy, both on the indicators of the NS and OS. After the therapy, the group of patients has a statistically significant decrease of MDA content, both in blood serum and in erythrocytes ($p<0.05$). At the same time, the analysis of the informative markers dynamics for antioxidant oxidative stress (AOS) of blood serum allowed to register a statistically credible increase in their mean values among patients after treatment ($p<0.05$): transferrin by 39.3% and SH-groups by 15.4%. Such changes in indicators contributed to the decrease in the intensity of the levels for the OS integral indicator, namely IOS almost 5 times less compared with the state before the complex therapy prescription ($p<0.001$). It should be noted that after the above treatment the patients had an increase in the indicators of SGA, SMC, and protein consumption with food ($p<0.05$).

Dynamics of NS parameters and OS markers among patients taking only intraperitoneal fluid with amino acids are presented in the table (Table 6). The results of the analysis allowed us to register no positive effect of the intraperitoneal fluid with amino acids on OS indicators for 3 months, except for transferrin content ($p<0.05$). However, the use of this fluid contributed to a significant improvement in serum albumin level ($p<0.001$). Thus, the results of the study show that the correction of NS disorders requires a comprehensive approach, which allows not only to carry out an adequate correction of ND but also to reduce the intensity of the LP processes.

Table 4: Comparative characteristics of end-stage renal disease patients in randomized groups.

Indicator	Groups of patients		P
	I group (n=20)	II group (n=20)	
Age (years, M±SD)	52.3±11.72	53.42±10.19	0.339
Men (n/%)	12/60	10/50	0.722
Duration of CAPD treatment (months, M±SD)	34.24±19.7	33.9±21.7	0.556
Type of kidney damage			
Non-diabetic glomerular lesions (n/%)	10/50	12/60	0.652
Non-glomerular lesions (n/%)	4/20	2/10	0.445
Diabetic nephropathy (n/%)	6/30	6/30	0.984
Nutritional parameters			
Serum albumin (g/l)	27.8±4.99	28.2±2.97	0.879
SGA (points)	3.09±1.83	1.88±0.41	0.698
BMI, kg/m^2, (m/w)	21.3±6.49/16.75±1.12	21.1±4.59	0.732
SMC, cm (m/w)	18.1±1.08/17.5±1.3	22.1±1.32/17.95±1.79	0.369
SFFT (skin and fat fold thickness), cm (m/w)	8.99±1.11/11.4±1.19	11.3±2.1/12.1±2.8	0.561

Discussion

Although peritoneal dialysis (PD) is a more biocompatible modality of dialysis compared to hemodialysis (HD), among patients treated with PD, oxidative stress intensity is likely to be higher compared with both the general population and patients withend-stage renal disease pre-dialysis stages [3-5]. We also demonstrated this assumption. It was stated that in the examined population of end-stage renal disease patients treated with CAPD the concentration of OS markers in comparison with the control group is increased.

During another study Xiao C, et al. (2018) having examined 64 patients treated with peritoneal dialysis, have proved that serum albumin level, a surrogate marker of nutritional disorders (ND), directly correlates with OS, it means patients with hypoalbuminemia have significantly higher OS levels indicators than patients with normal blood serum albumin content [19]. Other studies have confirmed his study as OS correction among patients with protein-energy malnutrition (PEM) contributes to the improvement of nutritional status (NS), namely the increase of serum albumin level, subjective global assessment (SGA), body mass index (BMI) and lean body mass [20-22]. Our conclusions during the investigation shows that OS

Table 5: Influence of Levocarnitine and intraperitoneal fluid with amino acids on OS and NS indicators among end-stage renal disease patients treated CAPD.

Indicator			Before treatment n=20	After treatment n=20	P
Blood serum content		Ceruloplasmin, g/l	0.302±0.09	0.315±0.109	0.572
		Transferrin, g/l	1.7±0.26	2.8±2.1	0.0001
		SH-groups	1.38±0.2	1.63±0.24	0.002
		IOS, rel.units	7.17±3.17	1.74±0.88	0.001
MDA	Serum	mcM/l	672.73±200.3	422.92±150.7	0.0055
	Erythrocytes		717.34±140.08	531.2±183.78	0.02
Albumin, g/l			27.8±4.99	29.3±4.53	0.49
SGA, points			3.09±1.83	4.80±1.63	0.049
BMI, kg/m^2			21.3±6.49	21.9±6.63	0.519
SMC, m/w. cm			18.1±0.08/17.5±0.03	20.3±0.16/19.8±0.09	0.034/0.027
SFFT, m/w. cm			10.99±1.11/13.4±1.19	11.31±1.14/13.1±1.56	0.745/0.998
Proteins, g/kg/round the clock			0.90±0.015	1.10±0.09	0.0004
Dietary calories, kcal/kg/day			31.8±0.76	32.34±1.2	0.283

indicators were found to be increased in all four groups, but they were the highest among the patients with moderate and severe malnutrition. The ceruloplasmin concentration in blood serum while comparing the groups did not change significantly. However, the content of MDA serum among patients with severe malnutrition was 2.36 times higher than in the group of patients with normal NS. Along with the increase in the production of MDA, in the group of patients with severe nutritional disorders there was revealed a decrease in the concentration of blood serum for transferrin in 2.4 times, SH-groups in 1.62 (62%) and ABC in 0.61 (46.8%) times compared with similar indicators of the group of patients with normal NS. At the same time, as severe malnutrition was present, an increase of IOS was found 4 times more compared with the group with normal NS. Many works show that it is important to have objective criteria for correcting O/A disorders to choose the optimal treatment regimen for these patients concerning inhibition of OS intensity. Thus, Wasserstein A (2019) in his study assumed that Levocarnitine is involved in most energy processes, its presence is obligatory for the oxidation of fatty acids, amino acids et cetera and used to increase muscle mass, thereby improving nutritional status indicators [23]. In the other hand, Taylor G (2012) investigated a fluid enriched with amino acids was developed for patients treated with PD and showed thatits main action is not only correction of PEM, but it can also be considered as a method of OS correction [24]. Our investigation showed that the prescription of complex therapy as a medicinal drugcontaining Levocarnitine together with intraperitoneal fluid with amino acids among end-stage renal disease patients treated with CAPD has a probable positive effect on the NS indicators SGA, SMC and protein consumption with food ($p<0.05$). Improvements in clinical indicators of NS are accompanied with possible positive dynamics of markers reflecting the balance of oxidative and antioxidant (O/A) reactions ($p<0.05$). Therefore, the results of the study have demonstrated that the concentration of OS markers is credibly higher among end-stage renal disease patients treated with CAPD compared with the control group ($p<0.05$). OS indicators have been proved to be increased in all four groups of patients with varying degrees of ND, however, they were the highest among patients with moderate and severe malnutrition. Credible strong feedback was registered between serum albumin and MDA blood serum levels ($r=-0.8830$; $p=0.0000$).

Conclusion

We found that the prescription of complex therapy as a medicinal drugcontaining Levocarnitine together with intraperitoneal fluid with amino acids among end-stage renal disease patients treated with CAPD has a probable positive effect on the NS indicators SGA, SMC and protein consumption with food. Improvements in clinical indicators of NS are accompanied with possible positive dynamics of markers reflecting the balance of oxidative and antioxidant reactions.It was stated that the usage of intraperitoneal fluid with amino acids once a day for 3 months had no positive effect on the decrease of oxidative stress indicators, except for the level of transferrin. The use of this fluid contributed to the improvement of the serum albumin level. Thus, this study concludes that it is necessary to adhere to the complexity of NS correction.

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