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ORIGINAL ARTICLE

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PARAMETERS OF FREE RADICAL OXIDATION AND ANTIOXIDANT DEFENSE IN PATIENTS WITH CERVICAL CANCER BEFORE AND AFTER RADICAL SURGICAL TREATMENT

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ABSTRACT

Purpose of the study. To analyse free radical oxidation and antioxidant defense in patients diagnosed with early cervical cancer (CC) before and after radical surgical treatment.
Patients and methods. Levels of diene conjugates, malondialdehyde (MDA), superoxide dismutase (SOD), catalase, glutathione, glutathione-dependent enzymes, vitamins A and E were determined in 74 women under the age of 45 (48 patients those who were at the stage of surgical treatment with a diagnosis of CC at the National Medical Research Center of Oncology in the period 2017–2020 and 26 healthy women).
Results. Patients with early CC showed significant changes in the intensity of lipid peroxidation processes and in antioxidant defense: elevate levels of MDA and diene conjugates, initial decline in the activity of SOD and catalase, low levels of vitamins A and E. These results complete the understanding of the processes occurring in the body of an oncological patient at the initial stage of tumor formation, which does not yet have an obvious clinical manifestation. After total removal of the ovaries, most of the indicators characterizing the enzymatic link of the antioxidant system tend to normalize, while the violation of the content of vitamins E and A (related to the non-enzymatic link of the antioxidant system) worsens.
Conclusions. Desynchronization of free radical oxidation processes with multidirectional changes in oxidation and antioxidation in patients with early CC at the stage of radical surgical treatment should be considered from the position of hormone-reducing surgery and a resulting complex of changes in the organs and systems of women with cancer.

Keywords: cervical cancer, lipid peroxidation, catalase, malondialdehyde, superoxide dismutase, glutathione

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Compliance with ethical standards: the ethical principles presented by the World Medical Association Declaration of Helsinki, 1964, ed. 2013 were observed in the study. The study was approved by the ethics committee of the National Medical Research Centre for Oncology (extract from the protocol of the meeting No. 5 dated 09/14/2019). Informed consent was received from all participants of the study.

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3.1.6. Онкология, лучевая терапия
ОРИГИНАЛЬНАЯ СТАТЬЯ

ПОКАЗАТЕЛИ СВОБОДНОРАДИКАЛЬНОГО ОКИСЛЕНИЯ И АНТИОКСИДАНТНОЙ ЗАЩИТЫ У ПАЦИЕНТОК С ДИАГНОЗОМ «РАК ШЕЙКИ МАТКИ» ДО И ПОСЛЕ ПРОВЕДЕНИЯ РАДИКАЛЬНОГО ХИРУРГИЧЕСКОГО ЛЕЧЕНИЯ

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РЕЗЮМЕ

Цель исследования. Проанализировать состояние свободнорадикального окисления и антиоксидантной защиты у пациенток с диагнозом «рак шейки матки» (РШМ) ранних стадий до и после проведения радикального хирургического лечения.

Пациенты и методы. Исследовали уровень диеновых конъюгатов, малонового диальдегида (МДА), супероксиддисмутазы (СОД), каталазы, глутатиона, глутатионзависимых ферментов, содержание витамина А и Е у 74 пациенток в возрастной категории до 45 лет (48 больных, находившихся на этапе хирургического лечения с диагнозом РШМ в ФГБУ «НМИЦ онкологии» Минздрава России в период 2017–2020 гг. и 26 здоровых женщин).

Результаты. У больных РШМ на начальных стадиях заболевания выявлены существенные изменения в интенсификации реакций перекисного окисления липидов и в антиоксидантной системе: повышенный уровень МДА и диеновых конъюгатов, исходное снижение активности СОД и каталазы, низкие показатели витамина Е и А. Данные результаты дополняют представления о процессах, происходящих в организме онкологического больного на начальном этапе формирования опухоли, которая ещё не имеет явного клинического проявления. После тотального удаления яичников большинство показателей, характеризующих ферментативное звено антиоксидантной системы, проявляют тенденцию к нормализации, в то время как нарушение содержания витаминов Е и А (относящихся к неферментативному звену антиоксидантной системы) усугубляется.

Заключение. Десинхронизацию процессов свободнорадикального окисления с разнонаправленными изменениями процессов окисления и антиокисления, у больных РШМ ранних стадий на этапе радикального хирургического лечения следует рассматривать с позиции гормонредуцирующей операции и связанного с ней сложного комплекса изменений в органах и системах женщин с онкологической патологией.

Ключевые слова: рак шейки матки, перекисное окисление липидов, каталаза, малоновый диальдегид, суперокиддисмутаза, глутатион

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Соблюдение этических стандартов: в работе соблюдались этические принципы, предъявляемые Хельсинкской декларацией Всемирной медицинской ассоциации (World Medical Association Declaration of Helsinki, 1964, ред. 2013). Исследование одобрено этическим комитетом ФГБУ «НМИЦ онкологии» Минздрава России (выписка из протокола заседания № 5 от 14.09.2019 г.). Информированное согласие получено от всех участников исследования.

Финансирование: работа проведена при поддержке ФГБУ «НМИЦ онкологии» Минздрава России.

Конфликт интересов: все авторы заявляют об отсутствии явных и потенциальных конфликтов интересов, связанных с публикацией настоящей статьи.

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RELEVANCE

The incidence of cervical cancer ranks 2nd (5.3 %) in the structure of oncogenital pathology in Russia, after uterine cancer (8.0 %) and 5th-6th in the structure of the entire oncopathology of the female population [1–3]. The tendency to increase the frequency of developing CC in women of reproductive age is alarming [4; 5]. The highest mortality rates from this pathology are recorded in the age group of 15–39 years (21.6 %) [6]. The increase in morbidity and mortality rates among young women diagnosed with CC [7] contributes to the active search for diagnostic and therapeutic directions with the effective integration of new scientific research. Modern trends in surgical treatment of oncogynecological patients are aimed at carrying out organ-preserving operations. According to the practical recommendations for the treatment of CC of localized stages, i.e. IA, IB1 and IIA1-2 from 2021, patients are shown to perform modified uterine extirpation (classification according to Piver III). Women diagnosed with CC under 45 years of age may retain ovarian function [8]. However, in some cases, if patients are not interested in preserving reproductive function, as well as in compliance with the principles of radical treatment, total removal of the affected organ and the most likely metastatic niches is used. Thus, the risk of implantation metastasis in ovarian cancer, the age of the patient and concomitant hormonal pathology determine the scope of the operation, i.e. extirpation of the uterus with appendages and expanded lymphadenectomy [9]. Forced and irreversible suppression of the ovaries leads to the formation of a pathological complex of disorders in the patient's body, which is due not only to the carcinolytic effects of the tumor process, but also to hypoestrogenia with a detailed metabolic picture of an artificially created menopausal state [10]. Postovariectomy syndrome (POES) manifests itself in the form of disorders of homeostasis, metabolic processes, neuroendocrine regulation and psychoemotional status, and is also characterized by changes in the state of pro- and antioxidant systems [9]. In addition, it is known that the growth and development of a malignant tumor, as well as the effect and negative manifestations of antitumor therapy largely depend on systemic disorders in the mechanisms of regulation of redox processes. During the formation of malignant transformation and the progression of the process, pathogenetically significant changes in the functional systems of the body of an oncological patient are generally recognized. There are results of studies where the role of oxidative stress and imbalance of redox processes in tumor growth is determined [11]. The activation of free radical oxidation (FRO) processes with the generation of activated oxygen metabolites in the regulation of cellular programs is proven [12]. There are studies that show significant changes in the level of the main antioxidant enzymes and vitamins A and E in patients with HPV (human papillomavirus) – associated forms of CC [13].

In order to improve the diagnosis and identify the patterns of development of CC, it is necessary to further study the pathophysiological changes occurring in the body of women of reproductive age at the early stages of the process and as a result of antitumor treatment. The results of research by domestic and foreign authors indicate that in the process of formation and progression of a malignant tumor, there is an increased activity of SRO with depletion of factors of the antioxidant system of the body of an oncological patient [14]. A number of key indicators of lipid peroxidation (LPO) and the activity of antioxidant protection of the blood of an oncological patient are informative in assessing the extent of the prevalence and progression of the tumor process. Long-term experimental and clinical studies conducted on the basis of the National Medical Research Center of Oncology allow us to characterize the state of redox reactions in most of the studied oncogynecological processes, which undoubtedly acquires practical significance [15; 16]. Thus, the basis of the method of treatment of patients with a primarily unresectable form of CC, including a combination of neoadjuvant chemotherapy and plasmapheresis procedure, were revealed violations among the indicators of free radical processes in endogenous intoxication in cancer patients at the stages of therapy [17]. At the same time, questions remain open about the dynamics of the indicators of LPO and the antioxidant system in patients with early-stage CC, which may be prognostically relevant in assessing the progression of the disease and the effectiveness of the antitumor treatment.

The aim of the study: to conduct a comparative analysis of the activity indicators of free radical oxidation and antioxidant protection processes in the blood of reproductive-age patients diagnosed with early-stage cervical cancer before and after radical surgical treatment.

PATIENTS AND METHODS

The blood parameters characterizing the state of activity of oxidative processes and antioxidant enzymes in 74 patients of reproductive age (up to 45 years) were studied. Of these, the main group consisted of 48 patients diagnosed with CC, hospitalized for complex treatment in the Department of Oncogynecology of the NMRC for Oncology of the Ministry of Health of Russia, and a comparison group of 26 women without oncopathology. The criterion for inclusion in the groups of this study was the absence of autoimmune and endocrine pathology, obesity, exacerbation of chronic pathology, as well as constant medication intake. The study was conducted with the approval of the Ethical Committee of the institution and with the voluntary consent of patients to use biological material with the processing of their personal data for scientific purposes. The main group is represented by patients diagnosed with CC, who underwent modified extirpation of the uterus with appendages (classification according to Piver III) as the first stage of antitumor treatment. According to the clinical examination, the distribution of the stage of oncological disease (classification TNM, 2019) is presented: $T_{1a2}N_0M_0$ in 5 patients (10.4 %), $T_{1b1}N_0M_0$ in 7 patients (14.7 %), $T_{1b2}N_0M_0$ in 36 patients (74.9 %). Morphological characteristics of the tumor: squamous cell carcinoma was detected in 40 patients (83.2 %), adenocarcinoma was diagnosed in 8 patients (16.8 %). A moderate degree of differentiation was detected in 27 patients (56.2 %). 97.4 % of patients had a positive HPV test of high carcinogenic risk. All patients of the group, in the early postoperative period, had signs of POES to one degree or another. The median age in the main group was 37 years, the average age was 41.6 ± 1.7, the range was 26–45 years. The body mass index (BMI) in the group was 23.8 ± 0.16 kg/m^2. Stages of the study: before surgical treatment and the 2nd day of the postoperative period. The comparison group consisted of 26 relatively healthy women, employees of the center, comparable to the group of patients with CC by age and BMI. The median age is 39 years, the average age is 43.2 ± 1.5, the range is 28–45 years. The BMI in the group without oncopathology was 23.3 ± 0.4 kg/m^2.

The intensity of LPO in erythrocytes and blood plasma was determined by the indicators of diene conjugates [18] and malondialdehyde (MDA) [19]. To judge the state of the antioxidant system, the activity of superoxide dismutase, catalase, glutathione-dependent enzymes, the level of glutathione [20], the content of vitamin A and E were determined by Chernyauskene R. Ch. method[21]. To determine these indicators, venous blood sampling from patients was carried out from the ulnar vein, in the morning, on an empty stomach.

Statistical results were processed using Statistika 10 programs according to the Student's criterion for two independent samples and the nonparametric Wilcoxon-Mann-Whitney criterion, the differences were considered statistically significant at $p < 0.05$. The samples were checked for compliance with the normal distribution according to the Kolmogorov-Smirnov criterion and the Shapiro-Wilk W-criterion. Since in most cases the distribution was close to normal, the results are presented in the form of $M \pm m$, where M is the sample mean, m is the error of the mean, the median (*Me*), which in all groups practically did not differ from M, and the interquartile spread in the form of counting the lower and upper quartiles: (Q25 and Q75).

RESEARCH RESULTS

The most informative integral indicators for assessing the intensity of LPO processes in the blood of patients are the content of primary products of diene conjugates and malondialdehyde (Table 1).

The status of antioxidant protection in patients with CC at the surgical stage of treatment was determined by several indicators: activity of catalase and superoxide dismutase enzymes, the level of reduced glutathione and glutathione-dependent enzymes (glutathione peroxidase, glutathione transferase, glutathione reductase). The data is shown in table 2.

Vitamin A and E levels in blood samples of CC patients before and after the surgery was the subject of special interest. The obtained data is brought in the table 3.

DISCUSSION

Malonic dialdehyde (MDA) is a secondary molecular product of LPO, formed in the body due to the reactions of ROS and polyunsaturated fatty acids. According to modern concepts, MDA is considered as a biological marker of changes in the system of free radical lipid oxidation [22]. When studying the initial activity of SEX processes in patients with a diagnosis of CC in comparison with healthy women, a significant increase in MDA was recorded, in blood plasma by 75.9 % (p = 0.002368), in erythrocytes by 59.7 % (p = 0.0108), the results are shown in Table 1.

According to our study, it was found that in patients with CC, the level of primary LPO products, which include diene conjugates, was significantly increased before the surgical stage of antitumor treatment. This indicator had significant differences with the group without oncopathology. So, in blood plasma, the indicator exceeded the values by 4.4 times (p = 0.000011), and in erythrocytes by an average of 6.6 times (p = 0.000038). However, a significant variation in the level of diene conjugates in erythrocytes was detected in the group of patients with CC, no changes were recorded in 42 % of cases, while a 14-fold increase in indicators was observed in 58 % of cases. After surgery, the level of diene conjugates in the group of patients with CC had a decrease of 5.3 times relative to the baseline level in erythrocytes, plasma, also recorded a decrease relative to the group of healthy and remaining 3.6 times higher than normal (p = 0.0033) (Table 1).

It is known that the main protective role under the influence of oxidative stress belongs to the non-enzymatic link of antioxidants (glutathione, vitamins A, E) and enzymatic antioxidants (superoxide dismutase (SOD), glutathione peroxidase, catalase) [23]. The results of the functioning of non-enzymatic and enzymatic antioxidant systems play an important role in the regulation of carcinogenesis, including in the processes that stimulate tumor progression, as well as the formation of its resistance to therapy [14].

Superoxide dismutase refers to the enzymes of the first line of defense of the antioxidant system.

Table 1. LPO indices in the blood plasma and erythrocytes in healthy women and CC patients before and after the surgery

Index *M ± m* Me (Q25; Q75)	Study group		
	Healthy women, *n* = 26	CC patients before treatment, *n* = 48	CC patients after surgery, *n* = 48
MDA in blood plasma (nM/ml)	7.075 ± 0.638 6.937 (4.9; 8.4)	12.476 ± 1.21 11.1 (4.245; 19.85) *p* = 0.002368	14.275 ± 1.424 14.1 (6.51; 19.4) *p* = 0.000567
MDA in 1 % erythrocyte hemolysate (nM/ml)	4.539 ± 0.363 4.54 (3.1; 5.651)	7.249 ± 0.734 5.5 (4.191; 7.25) *p* = 0.010766	7.564 ± 0.801 6.186 (4.409; 7.767) *p* = 0.008801
Diene conjugates in blood plasma (U/ml)	0.296 ± 0.041 0.21 (0.19; 0.37)	1.313 ± 0.156 0.9 (0.43; 2.29) *p* = 0.000011	1.055 ± 0.182 0.71 (0.355; 1.055) *p* = 0.003255
Diene conjugates in 20 % erythrocyte hemolysate (U/ml)	0.18 ± 0.028 0.12 (0.09; 0.22)	1.192 ± 0.168 0.76(0.14; 2.15) *p* = 0.000038 1) 0.171 ± 0.03 (20) 0.14 (0.1; 0.19) 2) 1.921 ± 0.192 (28) 1.9 (1.035; 2.7) *p* = 0.000000	0.229 ± 0.04 0.14 (0.1; 0.24) p^1 = 0.000000

Note: statistical significance of differences from parameters in healthy women – *p*, from parameters in CC patients before surgery – p^1.

Superoxide radicals directly activate the reactions of xenobiotic metabolism and prostaglandin synthesis, participate in the expression of certain genes and cell proliferation. A number of studies have shown that SOD, in addition to its antioxidant role, performs a regulatory function, while being the main link in ensuring a constant oxygen concentration. A change in superoxide dismutase activity can cause various pathological processes [24]. Thus, a decrease in the intensity of enzymatic reactions leads to insufficient protection from active forms of oxygen metabolites. And an increase in the activity of SOD contributes to the cytotoxic effect of hydrogen peroxide formed as a result of oxygen dismutation [14]. In our study, it was determined that the initial values of SOD in patients with CC compared with the value in the group of women without oncopathology had statistically significant differences. The activity of SOD erythrocytes in patients with CC was reduced by 30–31 % (p = 0.000000) both before treatment and after surgery in comparison with the "healthy" group. A plasma study of the total activity of SOD

Table 2. Parameters of the enzymatic unit of the antioxidant defense in the blood of healthy women and CC patients before treatment and after the surgery

Index *M ± m* Me (Q25; Q75)	Study group		
	Healthy women, *n* = 26	CC patients before treatment, *n* = 48	CC patients after surgery, *n* = 48
SOD in erythrocytes (act. U/ml)	110.79 ± 4.29 113.95 (101.1;125.9)	76.05 ± 3.12 72.16 (63; 89.8) *p* = 0.000000	77.54 ± 3.57 80.5 (56.6; 95.15) *p* = 0.000000
SOD in blood plasma (act. U/ml)	0.023 ± 0.002 0.022 (0.014; 0.032)	0.026 ± 0.0018 0.026 (0.015;0.033)	0.017 ± 0.0007 0.016 (0.012; 0.02) *p* = 0.000459 p^1 =0.000005
Catalase in erythrocytes (μM H_2O_2/min×mg Hb)	2673.8 ± 60.3 2720.5 (2500;2846.9)	2093.8 ± 131.2 2100 (1965.6;2491) *p* = 0.000069	2464.6 ± 129 25373 (2034.8;2689) p^1 = 0.052306
Catalase in blood plasma (μM H_2O_2/min)	52.02 ± 2.91 50.95 (42.01; 59.5)	58.11 ± 3.73 57.49 (46.25; 71.4)	69.62 ± 6.38 66.47 (52.22; 80.5) *p* = 0.010340
Reduced glutathione (μM/mg Hb)	35.81 ± 1.81 34.97 (27.96; 43.82)	32.67 ± 2.88 32.31 (23.61;38.05)	31.01 ± 2.42 29.86 (25.51; 40.05)
Glutathione reductase (IU/mg hemoglobin)	7.348 ± 0.94 7.095 (3.47; 10.97)	6.066 ± 0.758 5.28 (3.49; 7.69)	7.239 ± 1.053 7.385 (2.385; 10.23)
Glutathione peroxidase (IU/mg hemoglobin)	232.9 ± 23.7 218.6 (118.7; 344.4)	471.9 ± 42.9 447.7 (317.1; 624.7) *p* = 0.000005	380.5 ± 31.7 405.9 (307.8; 469.1) *p* = 0.000440 p^1 = 0.095117
Glutathione transferase (IU/mg hemoglobin)	69.97 ± 5.29 64.02 (49.03; 77.18)	61.35 ± 3.88 59.2 (52.74; 68.45)	61.15 ± 4.47 58.95 (54.18; 70.42)

Note: statistical significance of differences from indices in healthy women – p, from indices in CC patients before surgery – p^1.

revealed a decrease in patients with CC after surgery by 26.1–34.6 % ($p < 0.001$) compared with healthy women and the indicator for CC before treatment (Table 2).

Catalase is the main components of the first line of antioxidant protection, which, being an enzyme coupled with superoxide dismutase, decomposes hydrogen peroxide formed during the dismutation of the superoxide radical. The maximum accumulation of the enzyme was registered in erythrocytes. In the interstitial fluid of the body, catalase has no long-term activity, which is explained by the result of the action of proteolytic enzymes. It is believed that outside of erythrocytes, the enzyme does not have an obvious protective function. At the same time, a number of pathological conditions with the manifestation of the inflammatory process are characterized by an increased content of catalase, which provokes a decrease in the intensity of oxidation processes of functionally important structures [12]. In our study, catalase activity was analyzed in healthy patients and in patients with CC. Thus, in patients with CC, we revealed an initial decrease in catalase activity in erythrocytes by 21.9 % compared with the group of healthy women ($p < 0.001$) and an increase in indicators, and in plasma in the early postoperative

Table 3. Levels of vitamins E and A in the blood plasma and erythrocytes in healthy women and CC patients before treatment and after the surgery

Index *M ± m* Me (Q25; Q75)	Study group		
	Healthy women, *n* = 26	CC patients before treatment, *n* = 48	CC patients after surgery, *n* = 48
Vitamin E (erythrocytes) (U/ml)	0.177 ± 0.010 0.15 (0.14; 0.19)	0.266 ± 0.022 0.22 (0.17; 0.37) *p* = 0.002497	0.462 ± 0.037 0.470 (0.20; 0.73) *p* = 0.000000 p^1 = 0.000010
Vitamin E (plasma) (U/ml)	0.474 ± 0.011 0.465 (0.44; 0.54)	0.274 ± 0.019 0.27 (0.16; 0.36) *p* = 0.000000	0.346 ± 0.034 0.34 (0.22; 0.47) *p* = 0.003036 p^1 = 0.057561
Vitamin A (erythrocytes) (U/ml)	0.212 ± 0.010 0.20 (0.19; 0.26)	0.219 ± 0.022 0.13 (0.12; 0.40)	0.109 ± 0.010 0.115 (0.07; 0.15) *p* = 0.000000 p^1 = 0.000026
Vitamin A (plasma) (U/ml)	0.759 ± 0.022 0.78 (0.62; 0.87)	0.252 ± 0.023 0.210 (0.125; 0.37) *p* = 0.000000	0.104 ± 0.020 0.075 (0.05; 0.1) *p* = 0.000000 p^1 = 0.000007
E/A coefficient (erythrocytes)	0.978 ± 0.122 0.70 (0.538; 0.95)	1.461 ± 0.131 1.50 (0.405; 1.923) *p* = 0.012006	4.903 ± 0.519 3.286 (2.9; 6.635) *p* = 0.000000 p^1 = 0.000000
E/A coefficient (plasma)	0.635 ± 0.017 0.64 (0.59; 0.71)	6.013 ± 0.807 4.00 (3.00; 8.00) *p* = 0.000000	4.915 ± 0.390 5.00 (3.30; 5.80) *p* = 0.000000
Erythrocytes/plasma vitamin A ratio	0.383 ± 0.0285 0.333 (0.295; 0.422)	0.946 ± 0.074 0.881 (0.515;1.375) *p* = 0.000000	1.382 ± 0.097 1.353 (1.00; 1.851) *p* = 0.000000 p^1 = 0.098750
Erythrocytes/plasma vitamin E ratio	0.284 ± 0.014 0.321 (0.227; 0.333)	1.139 ± 0.107 1.00 (0.333; 2.00) *p* = 0.000000	1.496 ± 0.097 1.7958 (1.00; 2.00) *p* = 0.000000 p^1 = 0.016233

Note: statistical significance of differences from indices in healthy women – *p*, from indices in CC patients before surgery – p^1.

period by 24.4 % (p = 0.01) (Table 2). This suggests the release of an insignificant amount of the enzyme from erythrocytes into the blood plasma due to the possible destabilization of the membranes of blood cells as a result of surgical exposure.

The study of the indicators of glutathione and glutathione-dependent enzymes in the blood of patients with CC allows us to determine the activity of this line of the antioxidant system, with a possible prognosis of the course of cancer. Being an active link in the mechanism of the cellular antioxidant defense system, glutathione is considered as one of the main components in the pathophysiology of cancer [12]. The reduced form of glutathione in the form of tripeptide – Lyglutamyl-Lcysteinylglycine is present in cells of various types. In this compound, the presence of a sulfhydryl group and a gamma-glutamyl bond, being an electron donor, collectively determines the functions of glutathione as a reducing agent of nucleic acids, protein molecules and lipids. This property of the chemical compound makes it possible to significantly reduce the level of ROS in non-enzymatic and enzymatic reactions. Intracellular imbalance of reduced and oxidized glutathione is observed in a number of pathologies, including the development of a malignant process [25]. The data obtained by us on the content of glutathione in the studied groups did not reveal statistical differences, although its level was slightly lower in patients with CC before and after surgery. A comparative analysis of glutathione-dependent enzymes revealed certain changes. Thus, the activity of glutathione peroxidase (GPO) in patients with CC significantly exceeded the level in the group of women without oncopathology by 102.9 % (p = 0.0002). After surgery, the trend did not change, the GPO indicators in patients with CC remained elevated by 63.4 % (p = 0.003) relative to healthy women. The study of the dynamics of glutathione reductase and glutathione transferase indices, significant changes in the group of patients with CC compared with the group of healthy ones did not reveal (Table 2).

Thus, in the group of patients with CC, before the start of treatment, there was a more than twofold increase in the activity of GPO against the background of a statistically significant decrease in catalase activity in erythrocytes, which indicated a switch in the decomposition of hydrogen peroxide and organic hydroperoxides to non-toxic metabolites from the catalase pathway to the glutathione peroxidase pathway. After surgery, there was a partial restoration of the ratio between catalase and GPO, characteristic of women without oncopathology: the catalase/ GPO ratio was normal 11.5; in patients with CC before surgery 4.4; in patients with CC after surgery 6.5.

It should be borne in mind that glutathione-dependent enzymes perform an antioxidant function and are triggers in the regulation of peroxidation mechanisms. It is fair to note that the role of some enzymes in the formation of the malignant process is not unambiguous. Organic hydroperoxides are mediators of most physiological processes, actively participating in the regulation of cell proliferation and apoptosis. There are publications on the role of the enzyme glutathione peroxidase with the detection of its activation in squamous cell carcinoma cells and in lung adenocarcinoma [16].

It is known that the lack of vitamin E in the body leads to the destabilization of cell membranes, which leads to the breakdown of unsaturated fatty acids, as well as to a violation of their protein composition. Another equally effective antioxidant is vitamin A, which on the one hand interacts with free radicals, and on the other hand provides a constant level of vitamin E, thereby contributing to its antioxidant effect [14]. Prior to the start of antitumor treatment in the blood plasma of patients with CC, the initial values of vitamin E and A in comparison with the group of conditionally healthy were reduced by 41.9 % and by 74.4 % (p = 0.000000). The content of vitamin E in erythrocytes was increased by 50.3 % (p = 0.0025), and no significant changes were found for vitamin A. In the postoperative period, an increase in vitamin E indices was recorded in the erythrocytes of patients with CC by 2.7 times relative to women without pathology and by 1.8 times relative to the baseline level (p = 0.000000), while a decrease in the content of vitamin E in plasma was observed – by 27 % (p = 0.0030) relative to healthy. The level of vitamin A was reduced both in erythrocytes by 1.9 times and in plasma by 2.4 times (p = 0.000000) relative to healthy and by 2 and 2.4 times relative to the indicators before treatment (p < 0.0001). According to the results obtained, which generally do not contradict the literature data [26], the content of vitamin E in patients with CC is initially reduced only in blood plasma. At the same time, the values of the ratio of vitamins E and A (E/A) in

patients with CC of reproductive age before the start of antitumor treatment were statistically significantly increased relative to those in healthy women, with an increase in red blood cells by 1.6 times (p = 0.015) and 9.5 times (p = 0.011) in blood plasma. Data on changes in the E/A coefficient in oncogynecological patients are consistent with the results of previously published data. Thus, in the publication of E. M. Franzyants (1995), an increase of 2–5 times in the E/A coefficient in blood cells in patients with different tumor localization in the dynamics of radiation therapy was shown. When analyzing the results of the erythrocyte/plasma ratio (the ratio of the level of vitamins in erythrocytes to their plasma content), we revealed a significant increase in this coefficient for both vitamins relative to the values in healthy women, before surgery, the increase in the erythrocyte/plasma ratio for vitamin A was 2.5 times, for vitamin E 4 times (p = 0.000000). After the operation, a similar pattern was observed – the ratio of vitamins E and A in patients with CC exceeded 5.3 times in red blood cells, and in blood plasma by 7.7 times relative to the indicators in healthy women. The erythrocyte/plasma ratio in patients diagnosed with CC in the postoperative period had a statistically significant increase. The increase for vitamin E was 5.1 times, for vitamin A – 3.8 times (p < 0.0001). These changes in the erythrocyte/plasma coefficients for vitamins E and A were regarded by us as a manifestation of hyperpolarization of erythrocyte membranes in cancer patients diagnosed with CC. According to the literature, a recorded increase in the erythrocyte/plasma coefficients for vitamins A and E was described in patients with CC during chemotherapeutic exposure with no obvious clinical effect, which indicated the destabilization of erythrocyte membranes in this category of oncogynecological patients compared with a pronounced antitumor effect in patients with CC [26].

CONCLUSION

As a result of the study, it was found that in patients with CC at the initial stages of the disease, significant changes were detected in the intensity of the processes of LPO and in the antioxidant system. We found a significant increase in the level of MDA and diene conjugates, a decrease in the activity of SOD and catalase, at the same time, increased initial activity of GPO, as well as low levels of vitamin E and A. Undoubtedly, these results complement the understanding of the processes occurring in the body of an oncological patient at the initial stage of tumor formation, which does not yet have an obvious clinical manifestation. After total removal of the ovaries, most of the indicators characterizing the enzymatic link of the antioxidant system tend to normalize, while the violation of the content of vitamins E and A (related to the non-enzymatic link of the antioxidant system) worsens. Desynchronization of the processes of free radical oxidation with multidirectional changes in the processes of oxidation and antioxidation in patients with early-stage CC at the stage of radical surgical treatment should be considered from the perspective of hormone-reducing surgery and the associated complex of changes in the organs and systems of women.

References

1. Aksel EM, Vinogradova NN. Statistics of malignant neoplasms of female reproductive organs. Oncogynecology. 2018;3(27):64–78. https://doi.org/10.52313/22278710_2018_3_64, EDN: XZDJUD
2. Kaprin AD, Novikova EG, Trushina OI, Gretzova OP. The cervical cancer screening – unsolved problems. Research and Practical Medicine Journal. 2015;2(1):36–41. (In Russ.). https://doi.org/10.17709/2409-2231-2015-2-1-36-41, EDN: TOUJGB
3. Kit OI, Popova NN, Shikhlyarova AI, Frantsiyants EM, Moiseenko TI, Menshenina AP, et al. Development of postcastration syndrome and corrective effect of xenon in exponential dose regimen in young patients with gynecological cancers. South Russian Journal of Cancer. 2020;1(3):6–17. https://doi.org/10.37748/2687-0533-2020-1-3-1, EDN: DMUMOA
4. Bekhtereva SA, Domogirova AS, Vazhenin AV, Aksenova IA. Polineoplasia in patients with cervical cancer in the Chelyabinsk region of Russia. Research and Practical Medicine Journal. 2018;5(4):8–17. (In Russ.). https://doi.org/10.17709/2409-2231-2018-5-4-1, EDN: YQVQNN

5. Stuklov NI, Sushinskaya TV. Assessment of erythrocyte peripheral blood and activity of hemost asis in patients with cer vical cancer. Research and Practical Medicine Journal. 2016;3(1):17–23. (In Russ.). https://doi.org/10.17709/2409-2231-2016-3-1-2, EDN: VRNQSJ
6. Malignant neoplasms in Russia in 2020 (morbidity and mortality). Ed. by A. D. Kaprin, V. V. Starinsky, A. O. Shakhzadova. Moscow: P. A. Herzen MNIOI – Branch of the National Medical Research Radiological Center. 2021, 252 p. (In Russ.).
7. Senchukova MA, Makarova EV, Shurygina EI, Volchenko NN. Qualitative and quantitative characteristics of different types of tumor microvessels depending on the histological type of cervical cancer. Research and Practical Medicine Journal. 2020;7(4):36–50. (In Russ.). https://doi.org/10.17709/2409-2231-2020-7-4-4, EDN: ULSXFP
8. Khokhlova SV, Kolomiets LA, Kravets OA, Morkhov KYu, Nechushkina VM, Tyulyandina AS, et al. Practical recommendations for the drug treatment of cervical cancer. Malignant tumours. 2021;11(3s2-1):197–217. (In Russ.). https://doi.org/10.18027/2224-5057-2021-11-3s2-13, EDN: TLJDBM
9. Mazitova MI, Mardieva RR, Talipova IR, Antropova EYu. Climacteric syndrome. Clinical and epidemiological analysis. Russian Bulletin of Obstetrician-Gynecologist. 2021;21(5):66–72. (In Russ.). https://doi.org/10.17116/rosakush20212105166, EDN: IRLEHF
10. Sidorenko YuS, Kit OI, Popova NN, Arapova YuY, Shikhlyarova AI, Moiseenko TI, et al. The role of the central nervous system in inhibiting post-castration syndrome in patients with cervical cancer of reproductive age based on programmable xenotherapy regimens. Questions of Oncology. 2019;65(5):708–714. EDN: CGSWQU
11. Zaikina EV, Alliluev IA, Lazutin YuN, Przhedetskiy YuV, Shatova YuS, Engibaryan MA, et al. Investigation of the antioxidant status and the number of double-stranded DNA breaks in models of brain tumor lesion by metastases of non-small cell lung cancer in vivo. Research and Practical Medicine Journal. 2022;9(4):30–41. https://doi.org/10.17709/2410-1893-2022-9-4-3, EDN: PGKLSR
12. Andrisic L, Dudzik D, Barbas C, Milkovic L, Grune T, Zarkovic N. Short overview on metabolomics approach to study pathophysiology of oxidative stress in cancer. Redox Biol. 2018 Apr;14:47–58. https://doi.org/10.1016/j.redox.2017.08.009
13. Georgescu SR, Mitran CI, Mitran MI, Caruntu C, Sarbu MI, Matei C, et al. New Insights in the Pathogenesis of HPV Infection and the Associated Carcinogenic Processes: The Role of Chronic Inflammation and Oxidative Stress. J Immunol Res. 2018;2018:5315816. https://doi.org/10.1155/2018/5315816
14. Menshchikova EB, Zenkov NK, Lankin VZ, Bondar IA, Trufakin VA. Oxidative stress. Pathological conditions and diseases. Novosibirsk: Siberian University Publishing House. 2017, 284 p. EDN: QZDBCH
15. Surikova EI, Goroshinskaja IA, Nerodo GA, Frantsiyants EM, Malejko ML, Shalashnaja EV, et al. The activity of redox-regulatory systems in the tumor and its surrounding tissues in various histological types of tumor. Biomed Khimiya. 2016 Feb 2;62(2):187–192. https://doi.org/10.18097/PBMC20166202187, EDN: VUWAGF
16. Goroshinskaya IA, Surikova EI, Shalashnaya EV, Nerodo GA, Maksimova NA, Menshenina AP, et al. State of free radical processes in ovarian cancer with different prevalence and course of the disease. Izvestia University. The North Caucasus region. Series: Natural sciences. 2017;4(2):10–19. https://doi.org/10.23683/0321-3005-2017-4-2-10-19, EDN: YLSFHF
17. Menshenina AP, Zlatnik EYu, Sagakyants AB, Moiseenko TI, Ushakova ND, Franzyants EM, et al. New possibilities of immunocorrection in patients with cervical cancer in complex treatment. Russian Immunological Journal. 2021;24(1):115–122. https://doi.org/10.46235/1028-7221-373-NOO
18. Kopylova TN. A new method for the determination of conjugated dienes in blood serum. Cellular and Subcellular Experimental Pathology of the Liver. 1982, 135 c.
19. Andreeva LI, Kozhemyakin LA, Kishkun AA. Modification of the method for determining lipid peroxides in a test with thiobarbituric acid. Laboratory Business. 1988;(11):41–43. EDN: SKGPQJ
20. Arutyunyan AV, Dubinina EE, Zybina NN. Methods of evaluation of free radical oxidation and the antioxidant system of the body. Methodological Recommendations. St. Petersburg: Folio. 2000, 104 p.
21. Chernyauskene RCh, Varshkavyanichene ZZ, Grybauskas PS. Simultaneous fluorimetric determination of the concentration of vitamin E and A in blood serum. Laboratory Business. 1984;(6):362–365.
22. Gening TP, Fedotova AIu, Dolgova DR, Abakumova TV, Antoneeva II. The characteristics of redox-status of peripheral part of erythron under various localizations of neoplasm of organs of female reproductive sphere. Clinical laboratory diagnostics. 2017;62(8):468–472. https://doi.org/10.18821/0869-2084-2017-62-8-468-472, EDN: UEJRWX

23. Myandina GI, Hasan A, Azova MM, Tarasenko EV, Kulchenko NG. Influence of GSTP1 gene polymorhism on decreased semen quality. Russian Open Medical Journal. 2019;8(4). https://doi.org/10.15275/rusomj.2019.0411
24. Kulchenko NG. The main types of antioxidant therapy of pathospermia. Bulletin of the medical Institute "Reaviz": Rehabilitation, Doctor and Health. 2018;1(31):41–48. (In Russ.).
25. Goroshinskaya IA, Surikova EI, Frantsiyants EM, Neskubina IV, Nemashkalova LA, Medvedeva DE, et al. Redox forms of glutathione in malignant lesions of the stomach with varying aggressiveness degrees. Bulletin of Siberian Medicine. 2020;19(4):53–60. https://doi.org/10.20538/1682-0363-2020-4-53-60, EDN: RJRJZS
26. Franzyants EM, Sidorenko YuS, Rozenko LYa. Lipid peroxidation in the pathogenesis of tumor disease. Rostov-on-Don: Rostov University Press. 1995, 176 p.

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