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Nitrosative stress parameters as potential biomarkers in combat trauma

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As a result of severe injuries and post-traumatic stress disorder, sexual dysfunction and fertility disorders are among the complications men experience. The mechanisms of the effects of combat trauma are complex and include an imbalance of the immune system, which leads to severe inflammatory reactions and other immunomodifying effects after injury. An early response to an acute inflammatory injury, such as wound healing, is the production of nitric oxide (NO) as a result of L-arginine metabolism. NO is an important regulator of cellular functions throughout the wound healing process, stimulating fibroblasts to produce collagen, promoting matrix deposition, remodeling, and angiogenesis. However, insufficient or excessive NO synthesis negatively affects wound healing. The aim of the study was to investigate the prognostic power of arginase activity parameters and individual NO synthase isoforms as potential biomarkers of nitrosative stress in men with combat trauma. The study examined 68 men with combat trauma, including 42 men aged 20–39 years and 26 men aged 40–53 years. Criteria for inclusion in the control groups: 30 healthy men aged 20–39 years and 18 men aged 40–53 years with normal levels of cNOs, iNOs and arginase activity, somatically healthy, without sexual dysfunction. In all groups, the activity of NO synthases and arginase as markers of nitrosative stress was measured spectrophotometrically in blood serum and lymphocyte samples. The prognostic power of the parameters of cNOS, iNOS and arginase activity in the combat trauma and control groups was determined by the receiver operating characteristic curve (ROC curve). Based on the ROC analysis, the threshold value of cNOS activity in blood lymphocytes was determined, which is an integral highly sensitive criterion for unfavorable prognosis in combat trauma. For men aged 20–39 years, this figure is ≤37.5 nmol NADPH(H^+)/min. mg with a sensitivity of 61.9% and a maximum specificity of 100.0%, while for the group of men aged 40–53 years, the cutoff value is ≤38.4 nmol NADPH(H^+)/min. mg with a sensitivity of 65.4% and a maximum specificity. In the ROC analysis of iNOS activity in lymphocytes of men with combat trauma in relation to healthy men, an excellent model quality was obtained with the maximum area under the ROC curve for patients of both age groups. The lymphocyte arginase activity in the two age groups of men with combat trauma were characterized by the very good diagnostic accuracy of the test. Thus, the parameters of oxidative-nitrosative stress, in particular the activity of constitutive and inducible isoforms of NO synthase and arginase in blood serum and lymphocytes can be potential markers in distinguishing pathological changes in men affected by combat (bullet and shrapnel wounds). The inducible isoform of NO synthase has been shown in studies to be a highly sensitive and highly specific marker regardless of the age of men.

Keywords: combat trauma; blood lymphocytes; blood serum; arginase; NO synthase; ROC analysis.

Introduction

In armed conflicts of recent years, especially the Russian-Ukrainian war, the number of severe multiple and combined injuries has increased, mainly among male soldiers, due to the improvement of weapons and changes in combat tactics. Traumatic tissue damage leads to dysregulation of immune responses. The wounded suffer from complications such as slow wound healing, late onset of sepsis and infection, multiple organ dysfunction syndrome and acute respiratory distress syndrome. The mechanisms of combat trauma are complex and include an imbalance of the immune system, which leads to severe inflammatory reactions and other immunomodifying effects after trauma (Thompson et al., 2019). Severe injuries and post-traumatic stress disorder cause a variety of complications. Some of these complications are related to sexual health and fertility among men (Castillo et al., 2022; Nowak et al., 2023).

It has been established that oxidative-nitrosative stress plays a leading role in the pathogenesis of stress-induced disorders of the reproductive system (Satriano, 2004; Lundberg & Weitzberg, 2022; Reznikov, 2023; Kurhaluk & Tkaczenko, 2025). Nitric oxide is an important regulator of cellular functions throughout the wound healing process, stimulating fibroblasts to produce collagen, promoting matrix deposition, remodeling, and angiogenesis. Insufficient or excessive NO production negatively affects wound healing (Gould & Candy, 2017). Since the chemical and biological manifestation of NO effects depends on its concentration, the proximity of the biological target to the source of the regulator becomes critical. For example, cells or tissues close to macrophages, where high levels of NO are synthesized, will be exposed to direct and indirect effects through the primary and secondary effects of NO. On the contrary, if they are far from the NO source, they will experience only direct effects as the main mode of action of NO (Kiang, 2004; Lee et al., 2010).

L-arginine is an essential amino acid that is essential for the proper functioning of immune cells and plays an important role in many cellular processes such as ammonia detoxification, hormone secretion, and immune modulation (Racké & Warnken, 2010; Pedrazini, 2024). The pathways of L-arginine metabolism in inflammation include its conversion to the cytostatic molecule NO, and the breakdown of L-arginine to urea and ornithine by arginase. These pathways are subject to temporal regulation in models of acute inflammation, such as wound healing and glomerulonephritis. L-arginine is a precursor of citrulline, symmetrical and asymmetrical guanidine, L-homoarginine, methylene homologue, and NO (Satriano, 2004; Pedrazini et al., 2024). The latter is a key component of endothelial relaxation factor, an endogenous messenger molecule that has endothelium-dependent physiological effects on the cardiovascular system. Arginase I plays a major role in the urea cycle by catalyzing the cytosolic hydrolysis of L-arginine to ornithine and urea. Studies have shown that human liver arginase I can be an effective drug in the enzymatic therapy of certain types of cancer by inducing arginine starvation of malignant tumor cells (Stasyuk et al., 2017).

Under normal physiological conditions, NO exerts anti-inflammatory effects, but in pathological conditions, its excessive production acquires pro-inflammatory properties (Sharma et al., 2007). Indu-

cible NO synthase (iNOS), a calcium-independent enzyme, is mainly expressed in macrophages and tissues in response to (pro)inflammatory mediators (Qualls et al., 2012). Since L-arginine is a substrate for NO synthase, amino acid derivatives are becoming important biomarkers in clinical diagnostics in determining the relationship between their levels and the course of the disease, in particular in kidney and liver disease, cardiovascular disease (Tsikas & Wu, 2015; Stasyuk et al., 2017). The amount of NO produced depends on the activity of individual NOS isoforms. Increased expression of iNOS is a key effector mechanism that is triggered early after injury and is usually associated with the immune system. NO production by iNOS occurs over a long period of time (Thomas et al., 2008; Wink et al., 2011). Infiltrating macrophages secrete either proinflammatory cytokines or anti-inflammatory cytokines with increased arginase activity, which is apparently crucial for the regulation of subsequent phases of wound healing. The products of active iNOS and arginase metabolic pathways regulate each other through feedback (Gould & Candy, 2017). When both iNOS and arginase enzymes are induced together, peroxynitrites formed under arginine limitation conditions cause apoptosis of activated T lymphocytes (Bronte et al., 2003).

NO provides cytoprotection generated by iNOS, which is expressed in response to bacterial/inflammatory stimuli, and plays an important role in the wound healing process. In addition, NO has antimicrobial and cytotoxic properties that are necessary to maintain sterility in the wound space (Gould, & Candy 2017; Angka et al., 2022). Patients with combat injuries demonstrate changes associated with cytokines as proinflammatory mediators that activate iNOS and inducible cyclooxygenase (Li et al., 2024). Regardless of the age of patients with combat trauma, iNOS activity has been shown to be a highly specific and highly selective marker in blood serum and lymphocytes (Onufrovych et al., 2024). An increase in iNOS activity in patients with a mixed form of erectile dysfunction in combat trauma was accompanied by a corresponding decrease in cNOS activity (Vorobets et al., 2024).

It is believed that cNOS produces low concentrations of NO in the submicromolar range for limited periods of time, which causes a wide range of physiological effects. On the contrary iNOS generates NO for a long period of time in local concentrations up to 1–5 μM and causes pathophysiological effects (Kiang, 2004; Musci et al., 2006). Traumatic injury is characterized by the development of L-arginine deficiency due to increased catabolism of this amino acid (Marik & Flemmer, 2012; Canè et al., 2024). It has been shown that after surgical trauma, the amount of L-agrinine decreases rapidly (Angka et al., 2017). Decreased levels of L-arginine following traumatic stress or injury can promote wound healing through arginase activity, which promotes the formation of polyamines and proline, which enhance collagen synthesis and wound closure. Pathways involving L-arginine and the urea cycle are critical for wound healing.

Arginase and NOS are enzymes that catalyze multi-enzyme reactions that synthesize physiologically important products depending on the concentration of L-arginine (Stasyuk et al., 2017). Arginine activity of lymphocytes is an important marker indicating a humoral response of the immune system to an antigen. A significant increase in arginase activity has been found in inflammatory processes caused by staphylococci (Lavryk, 2017). The plasma of healthy individuals usually has low levels of arginase, but under certain pathological conditions its activity may increase (Morris, 2012). Arginase ensures the formation of proline and ornithine from L-arginine for collagen deposition and polyamine accumulation, respectively (Gould & Candy, 2017). An increase in arginase activity is necessary to promote wound healing. If enzyme activity is inhibited, inflammation in the wound increases, impairing matrix formation and delaying wound healing (Campbell et al., 2013). The literature shows that the activity of NO synthases changes during aging (Sverdlov et al., 2014; Toma et al., 2021). Therefore, the study of the prognosticity of nitrosative stress indicators was carried out for two age groups of men affected by combat operations (bullet and shrapnel wounds).

The aim of present study was to investigate the prognostic power of arginase and NO synthase isoforms as potential biomarkers of nitrosative stress in men with combat trauma.

Materials and methods

The study examined 68 men with combat trauma, including 42 men aged 20–39 and 26 men aged 40–53. Inclusion criteria for the control groups: 30 healthy men aged 20–39 years and 18 men aged 40–53 years, somatically healthy, without sexual dysfunction, cardiac, endocrinological and neurological pathology. The study was carried out in compliance with the principles of medical ethics and protection of patients' rights, human dignity and moral and ethical standards, in accordance with the principles of the Helsinki Declaration of Human Rights, the Council of Europe Convention on Human Rights and Biomedicine, and the legislation of Ukraine; the study received the permission of the Bioethics Committee of Danylo Halytskyi Lviv National Medical University (protocol No. 7 of 26.06.2023).

In all groups, the activities of NO synthases and arginase as markers of nitrosative stress were measured spectrophotometrically in blood serum and lymphocyte samples. The prognostic power of the parameters of cNOS, iNOS and arginase activity in combat trauma and control groups was determined by the receiver operating characteristic curve (ROC curve). The main application of ROC analysis is to determine the predictive ability of biomarkers for diagnostic and prognostic purposes. Generally, a biomarker is defined as a laboratory measurement that reflects the activity of a disease process. Determining the optimal cutoff point for biomarkers in diagnostic studies is one of the tasks of ROC analysis (Katz, 2004; Hassanzad & Hajian-Tilaki, 2024).

A receiver operating characteristic curve, or ROC curve, is a visual tool for assessing the accuracy of a classifier or predictor. The ROC curve is a plot of sensitivity versus (1-specificity) and indicates the performance of the test. The area under the ROC curve (AUC) is a common summary statistic of diagnostic accuracy. ROC curves for diagnostic tests with perfect discrimination between negative and positive reference samples pass through the coordinates (0;1), which represent 100% Se (sensitivity) and Sp (specificity). In this case, the test is considered perfect. The classification of accuracy by AUC for a diagnostic test is as follows: poor ($0.5 < AUC < 0.6$), average ($0.6 < AUC < 0.7$), good ($0.7 < AUC < 0.8$), very good ($0.8 < AUC < 0.9$), excellent ($0.9 < AUC < 1$). For the AUC value, a confidence interval (CI) with a probability of 95% was calculated, which is presented in the form of lower and upper limits of the CI (Swets, 1988; Greiner et al., 2000). The value of the optimal cutoff point is defined as the mean value plus two standard deviations of the negative standard sample. Sensitivity, specificity, and area under the ROC curve were calculated with standard error (SE) and 95% CI. The threshold of significance at $P < 0.05$ is considered significant, at $P < 0.001$ – highly significant, and statistically insignificant or unreliable at $P > 0.05$. Statistical processing of the results and diagrams were performed using the MedCalc software package® Statistical Software version 22.018 (MedCalc Software Ltd, Ostend, Belgium; www.medcalc.org).

Results

ROC analysis, which is a graph of the dependence of the sensitivity of an indicator on the relative number of false-negative results, was used for a qualitative analysis of the diagnostic significance of biomarkers: activity of constitutive isoform of NO synthase, activity of inducible isoform of NO synthase, serum and lymphocytes arginase activity in men affected by combat. ROC curves were constructed for each enzyme to determine the ability of oxidative-nitrosative stress indicators to distinguish pathology in combat trauma. The results of the quantitative evaluation of the constructed curve by calculating the AUC area for cNOS activity in the blood serum and lymphocytes of men with military combat trauma are shown in Table 1. The characteristic ROC curves for serum cNOS activity for two age groups of men are stretched towards the upper left corner, i.e., they are characterized by a large area (Fig. 1). The analysis of ROC curves indicates good diagnostic value of serum cNOS activity for men of both age groups. The data in Table 1 show that serum cNOS activity is characterized by moderate test accuracy for men

aged 20–39 years and is highly sensitive and moderately specific for the age group of 40–53 years. The cut-off point for serum cNOS activity was set at ≤ 4.2 nmol NADPH(H^+)/min. mg with a sensitivity of 78.6% and specificity of 80% for men aged 20–39 years. With a high sensitivity of 96.2% and moderate specificity of 66.7%, the optimal cut-off point for serum cNOS activity was ≤4.1 nmol NADPH(H^+)/min-mg protein for men aged 40–53 years.

Table 1
ROC analysis of constitutive NO synthase activity in men with military combat trauma (N = 68)

Indicator for cNOS activity	Blood serum		Lymphocytes	
	20–39 years old	40–53 years old	20–39 years old	40–53 years old
Se, %	78.57	96.15	61.90	65.38
Sp, %	80.00	66.67	100.00	100.00
AUC	0.832	0.752	0.773	0.819
95 % CI AUC	0.725–0.910	0.599–0.870	0.660–0.864	0.674–0.919
P	<0.0001	0.0014	<0.0001	<0.0001
Standard error	0.0481	0.0791	0.0554	0.0629
Associated criterion	≤4.2	≤4.1	≤37.5	≤38.4

Note: Se – sensitivity; Sp – specificity; AUC – area under the curve; 95% CI AUC – 95% confidence interval for area under the curve; P – significance level compared to AUC.

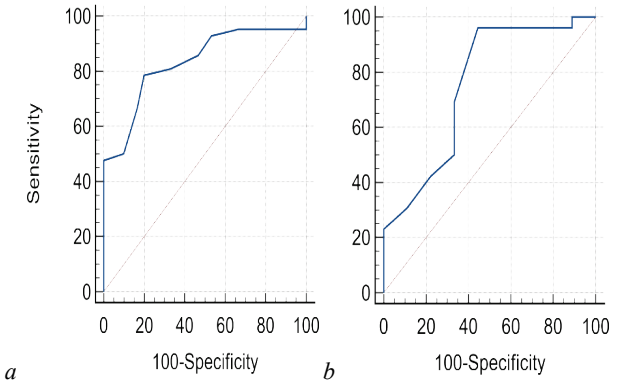

Fig. 1. Diagnostic value of serum cNOS activity in men with military combat trauma versus the control group: *a* – 20–39 years; *b* – 40–53 years; N = 68

ROC curve plots for the activity of constitutive NO synthase in lymphocytes of the examined men are located as far as possible to the upper left corner for the two age groups (Fig. 2).

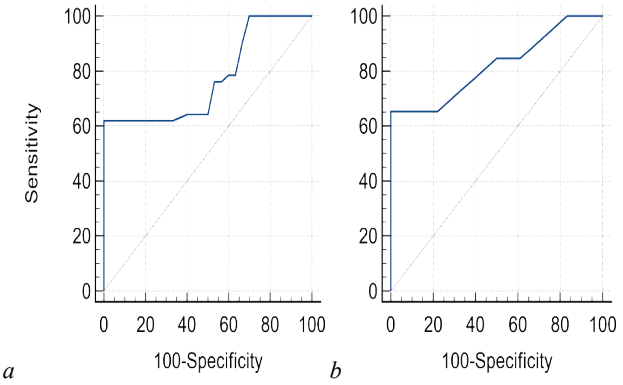

Fig. 2. Diagnostic value of lymphocytes cNOS activity in men with military combat trauma versus the control group: *a* – 20–39 years; *b* – 40–53 years; N = 68

The estimation of the ROC curve for detecting pathological changes in men with combat trauma by the cNOS activity in blood lymphocytes assesses the marker as having good diagnostic value for two age groups.

Based on the ROC analysis, the threshold value of cNOS activity in blood lymphocytes was determined. It is an integral highly sensitive criterion for unfavorable prognosis in men with military combat trauma. The cutoff value of cNOS activity was ≤ 37.5 nmol NADPH(H+)/min. mg with a sensitivity of 61.9% and a maximum specificity of 100% for men aged 20–39 years. The cutoff value was ≤38.4 nmol NADPH(H+)/min. mg with a sensitivity of 65.4% and a maximum specificity for the group of men aged 40–53 years.

The results of the quantitative evaluation of the constructed curve by calculating the AUC area for iNOS in blood serum and lymphocytes are shown in Table 2. In the ROC analysis of iNOS activity in the blood serum of men with combat trauma in relation to healthy men, an excellent diagnostic quality of the model with the maximum area under the curve was obtained (Fig. 3).

Table 2
ROC analysis of inducible NO synthase activity in men with military combat trauma (N = 68)

Indicator for iNOS activity	Blood serum		Lymphocytes	
	20–39 years old	40–53 years old	20–39 years old	40–53 years old
Se, %	100.00	100.00	100.00	100.00
Sp, %	100.00	100.00	100.00	100.00
AUC	1.00	1.00	1.00	1.00
95 % CI AUC	0.950–1.000	0.920–1.000	0.950–1.000	0.920–1.000
P	<0.0001	<0.0001	<0.0001	<0.0001
Standard error	0.0390	0.000	0.000	0.000
Associated criterion	>0.3	>0.3	>2.3	>1.9

Note: see Table 1.

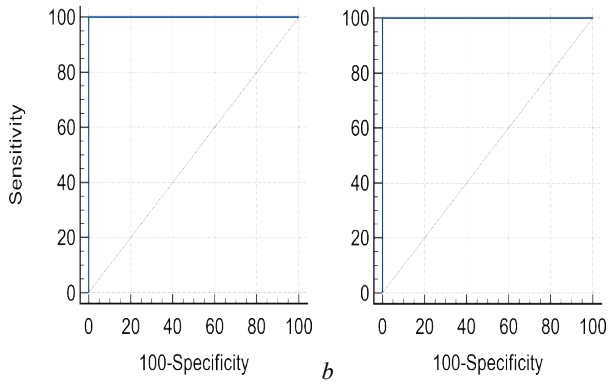

Fig. 3. Diagnostic value of serum iNOS activity in men with military combat trauma versus the control group: *a* – 20–39 years; *b* – 40–53 years; N = 68

The optimal cutoff point for iNOS activity in the serum of men aged 20–39 years to determine a positive test for detecting pathology in combat trauma was established at the level of >0.3 nmol NADPH(H^+)/min-mg protein at the maximum values of sensitivity and specificity. The threshold value for men aged 40–53 years was obtained at the same level. This value of the marker is highly specific and highly selective for both age groups. In the ROC analysis of iNOS activity in blood lymphocytes of men with combat trauma relative to healthy men, an excellent model quality was obtained with the maximum area under the ROC curve for patients of both age groups (Fig. 4).

The optimal cut-off point iNOS activity for the age group 20–39 years was >2.3 nmol NADPH(H+)/min. mg at the maximum values of specificity and sensitivity. The ratio of probability of negative test results was almost perfect <0.001. ROC-analysis of lymphocyte iNOS activity for the age group of 40–53 years also gave excellent ROC curve results. The optimal cutoff point was >1.9 nmol NADPH(H^+)/min. mg protein determined by the maximum values of sensitivity and specificity. The enzyme is characterized by high specificity and sensitivity.

The results of the prognostic ability of arginase activity in blood serum and lymphocytes in men with combat trauma are shown in Table 3. In the ROC analysis of serum and lymphocyte arginase activity, good quality models were obtained for both age groups (Fig. 5). The optimal cut-off point for serum arginase activity was determined to be ≤ 11 nmol urea/min. mg protein in men aged 20–39 years, which corresponds to a good specificity value of 86.7% and

sensitivity of 73.8%. The optimal cut-off point for serum arginase activity was determined by ≤ 4.1 nmol urea/min. mg protein for men aged 40–53 years, which is characterized by a high sensitivity value of 96.2% and an average specificity value of 55.6% with an almost perfect ratio of probability of a positive test result P < 0.0001.

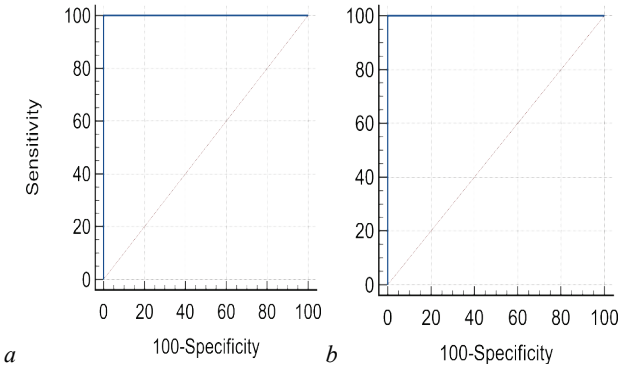

Fig. 4. Diagnostic value of lymphocytes iNOS activity in men with military combat trauma versus the control group: *a* – 20–39 years; *b* – 40–53 years; N = 68

The areas under the ROC curves for lymphocyte arginase activity in two age groups of men with combat trauma were determined. They were characterized by the very good diagnostic accuracy of the test (Fig. 6). The optimal cut-off point for lymphocytes arginase activity iwa ≤ 84.4 nmol urea/min per mg protein for the age group 20–39 years with a maximum sensitivity and average specificity of 63.3%. This value was ≤38.4 nmol urea/min per mg protein, obtained with a maximum specificity of 100% and an average sensitivity of 65.4% for men aged 40–53 years (Table 3).

Table 3
ROC analysis of arginase activity in men with military combat trauma, N = 68

Indicator for arginase activity	Blood serum		Lymphocytes	
	20–39 years old	40–53 years old	20–39 years old	40–53 years old
Se, %	73.81	96.15	100.00	65.38
Sp, %	86.67	55.56	63.33	100.00
AUC	0.796	0.752	0.822	0.819
95 % CI AUC	0.685–0.882	0.599–0.870	0.714–0.902	0.674–0.919
P	<0.0001	0.0014	<0.0001	<0.0001
Standard error	0.0532	0.0791	0.0558	0.0629
Associated criterion	≤11	≤4.1	≤84.4	≤38.4

Note: see Fig. 1.

Discussion

The study highlights the possibility of using the parameters of oxidative-nitrosative stress as biomarkers of pathological changes caused by military combat trauma. Injuries as a result of hostilities (bullet and shrapnel) with the use of modern weapons in 35% of cases are combined/multiple or combined, which requires rapid decision-making related to the provision of medical care. Combat trauma is characterized by a specific course of the wound process and high-risk clinical pathological manifestations that directly threaten the safety of a soldier's life (Lee et al., 2001; Dufour-Gaume et al., 2023; Gumeniuk & Korol, 2024).

The pathogenesis of combat trauma complications occurs at the systemic, organ, cellular, subcellular, and molecular levels (Makarov et al., 2024). From the molecular point of view, the study of pathogenesis has a great advantage in the diagnosis and treatment of diseases, allowing the progression of the disease to be assessed based on changes at the molecular level (Lee et al., 2018). One of the physiological responses to combat trauma is the activation of the peripheral immune system, which is caused by mechanical injuries: shrapnel and bullet wounds. The consequences of combat trauma are associated with a unique set of protein biomarkers (Chromy et al., 2013) that can be used to diagnose, predict or prognosticate pathological conditions.

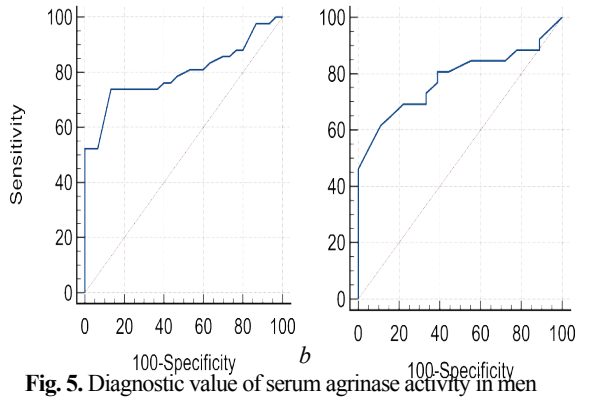

Fig. 5. Diagnostic value of serum agrinase activity in men with military combat trauma versus the control group: *a* – 20–39 years; *b* – 40–53 years; N = 68

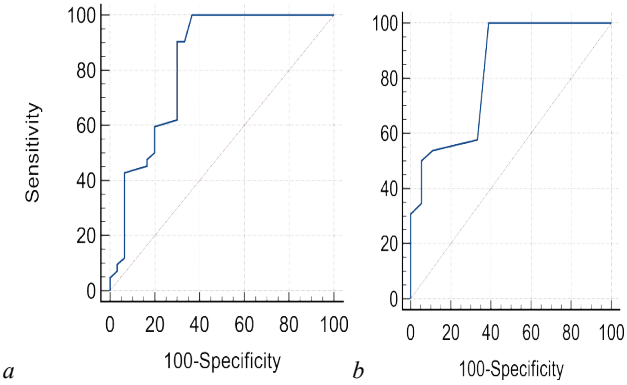

Fig. 6. Diagnostic value of lymphocytes agrinase activity in men with military combat trauma versus the control group: *a* – 20–39 years; *b* – 40–53 years; N = 68

Determining the optimal thresholds for quantitative biomarkers plays a crucial role in clinical decision-making in diagnostic medicine (Hassanzad & Hajian-Tilaki, 2024). ROC analysis is an additional choice for determining the optimal threshold value. In this study, we used the Youden method to obtain the optimal cutoff point. In its clinical interpretation, this criterion maximizes the proportion of correct classification after adjusting for randomness. In present study, the optimal cutoff points were obtained for the activity of constitutive and inducible isoforms of NO synthases and arginase in blood serum and lymphocytes.

Results on the clinical trial of biomarkers in men with combat trauma showed that as we move from the lower left corner to the upper right corner, the sensitivity increases and the specificity decreases. As a result, the cutoff value gets lower and lower, and at the end of the upper right corner of the square, the sensitivity and specificity are 1 and 0, respectively, which is the lowest possible cutoff value for this test. In the study, iNOS activity in blood serum and lymphocytes of two age groups of men were determined to be highly sensitive and highly specific indicators of oxidative-nitrosative stress in combat trauma in military personnel. AUC is one of the indicators of diagnostic accuracy when comparing diagnostic tests in ROC analysis. The area under the ROC curve is 1 for iNOS activities in both blood serum and lymphocytes, indicating its maximum values and the ideal predictive value. In contrast, serum cNOS activity as a test is of medium specificity and very good sensitivity for the age group 40–53 years. The test is defined as of medium sensitivity and good specificity for the group of men aged 20–39 years. The cNOS activity as a test for detecting pathological changes in combat trauma performed on lymphocytes is of medium sensitivity but highly specific. The area under the ROC curve is in the range of 0.7–0.9, which also indicates a good predictive ability of the test.

Serum arginase activity as a marker value is characterized by good sensitivity and specificity for men aged 20–39 years. This marker for men of the second age group is highly sensitive and low specific. Agrinase activity of lymphocytes is characterized by excellent

sensitivity and low specificity for men aged 20–39 years. Compared to the first age group of men, the data on arginase activity for the second group of men is characterized by good sensitivity and excellent specificity. The area under the ROC curve within the defined limits indicates that arginase activity has good prognostic value. Thus, our study demonstrates that the specificity and sensitivity of serum and lymphocyte iNOS activities are high for predicting pathological changes in men with military combat trauma of different ages.

At lower values of cNOS activity than the determined optimal cutoff points, with high sensitivity and specificity, pathological processes associated with combat trauma are likely to occur. The results are consistent with our previous studies (Vorobets et al., 2024). At the same time, higher values of iNOS activity as the optimal cutoff point with high sensitivity and specificity indicate inflammatory or pathological processes, which is supported by the literature showing the increase in iNOS activity in the acute and early course of the wound process and infectious diseases (Gould & Candy, 2017; Angka et al., 2022). The optimal cutoff point for arginase for both serum and lymphocytes for men of both age groups was determined, indicating that a value lower than this probably indicates inflammatory or pathological processes associated with combat trauma. Obviously, the arginase activity, which provides hydrolysis of L-arginine to ornithine and urea, decreases in inflammatory processes, and L-arginine as a substrate for which arginase and NO synthases compete is metabolized by iNOS into NO. In acute wounds, iNOS activity predominates at the early stage of inflammation, where the wound microenvironment is cytotoxic (Angka et al., 2017).

The ROC analysis established the parameters that have the greatest diagnostic significance for enzyme indicators, which can be used as additional criteria for establishing pathological changes in men with military combat trauma.

Conclusion

Parameters of oxidative-nitrosative stress, in particular, the activity of constitutive and inducible isoforms of NO synthase and arginase in blood serum and lymphocytes, can be potential markers in distinguishing pathological changes in men with military combat trauma (bullet and shrapnel wounds). The inducible isoform of NO synthase has been shown to be a highly sensitive and highly specific marker regardless of the age of men. The optimal cut-off points for the diagnostic parameters of the constitutive isoform of NO synthase, inducible isoform of NO synthase and arginase at the maximum values of sensitivity and specificity in the blood serum and lymphocytes of men of two age groups were found. Determination of the activity of NO synthase isoforms and arginase is important in detecting pathological changes in men with military combat trauma.

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