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METHOD OF CORRECTION OF ENDOTHELIAL DYSFUNCTION WITH COMBINATION OF ADEMETIONINE AND TAURINE

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Abstract. In the experiment, was made a simulation of endothelial dysfunction in male rats of Wistar-line byintraperitoneal administration of L-NAME at a dose of 25 mg/kg/day for 7 days. Deficiency of nitric oxide in result of the blockade of NO-synthase was accompanied by violation of endothelium-dependent and endothelium-undependentvasodilation assessed in pharmacological trials, which was reflected in the increase of the coefficient endotelialny dysfunction.In this case, for correction of endothelial dysfunction intraperitoneallyademetionine in the dose of 150 mg/kg and after an hour taurine at a dose of 260 mg/kg was injected the animal once a day for 7 days.
The method provides effective impact of the combination of hepatoprotectorAdemetionine in the dose of 150 mg/kg/day and a sulfur-containing amino acid Taurine in the dose of 260 mg/kg/day on the functioning of the vascular endothelium, and has endotheliopathy effect on models L-NAME-induced deficiency of nitric oxide, which includesthe endothelium-dependent vasodilation and the decrease of coefficient of endothelial dysfunction.
Keywords: *L-NAME*, endothelial dysfunction, taurine, ademetionine, nitric oxide, oxidative stress.

Intro: As already known, the endothelium maintains homeostasis by regulating the balance of opposing processes: vascular tone, responsible for vasodilation and vasoconstriction; anatomical structure of blood vessels, regulating the synthesis and the inhibition factors of cell proliferation; hemostasis, participating in the synthesis and inhibition of fibrinolysis factors and platelet aggregation; local inflammation by producing Pro - and anti-inflammatory cytokines [1, 2, 3, 4]. The endothelium lines all vessels, regardless of their organ localization, so endothelial dysfunction, the basis of which is reduced synthesis of endothelial cell nitric oxide (NO) is a predictor of diseases, not only arteries and veins, but components of the microvasculature [5, 6]. Endothelial dysfunction explains the pathogenesis of diseases such as hypertension, atherosclerosis, coronary artery disease, cardiomyopathy, pathogenesis of chronic heart failure, metabolic disorders: hyperlipidemia, hyperhomocysteinemia, hypoestrogenemia, diabetic vascular lesions, venous transformation [7, 8, 9, 10]. Therefore, the goal of our research was the search of drugs, their combinations are capable of correcting endothelial dysfunction, having endotheliopathy effect.From the literature it is known that taurine has antihypertensive, antioxidant [11] activities, contributes to endothelium-independent vasodilatation [12], the lack of endogenous taurine inhibits the processes of vasorelaxation[13].

Endothelial dysfunction is a prerequisite for the development of atherosclerosis.

It is known that under the action of ademetionine normalization coefficient of endothelial dysfunction and that treatment with S-adenosyl-l-methionine (SAM) prevents endothelial dysfunction in animals by induction of hemoxygenase-1 (HO-1) in endothelial cells of blood vessels and I believe that treatment (CAM) may represent a new therapeutic strategy for atherosclerosis [14, 15].

Main part: In connection with the foregoing, the purpose of this study was the investigationthe endotheliopathy properties of a combination of ademetionine in the dose of 150 mg/kg/day and taurine at a dose of 260 mg/kg/day as one of the possible effective combinations with dysfunction of endothelium, L-NAME-induced [16, 17, 18] deficiency of nitric oxide.

Materials and methods research. Experiments were carried out on white rats-males of Wistar-line

weighing 170-220 g. (n=10 animals). L-NAME was injected intraperitoneally at a dose of 25 mg/kg/day. Ademetionine and taurine and their combination are entered daily intragastrically (by gavage) at doses of 150 mg/kg/day and 260 mg/kg/day accordingly, at intervals of an hour, for 7 days.On the eighth day from the beginning of the experiment under combined anesthesia (chloralgidrate 150 mg/kg and zoletile 60 mg/kg) was injecteda catheter in the left carotid artery for registration of blood pressure (BP), vascular samples was carried out by introduction of a bolus into the left femoral vein of pharmacological agents.Hemodynamic parameters: systolic blood pressure (SBP), diastolic blood pressure (DBP) and heart rate (HR), measured continuously by a sensor TSD104A and hardware-software complex MP100, production Biopac System, Inc., USA.Functional tests: endothelium-dependent vasodilation the (EDV) – intravenous administration of acetylcholine (AH) at a dose of 40 mg/kg, endothelium-independent vasodilation (EIV) – intravenous injection ofnitroprusside sodium (NP) at a dose of 30 µg/kg.

To assess the degree of endothelial dysfunction in experimental animals and its correction by researched drugs we have performed the calculation of the coefficient of endothelial dysfunction (CED) [16, 17, 18, 19].

With statistical data processing was calculated average value, standard deviation. The differences were considered significant at $p<0.05$. The normality of distribution was checked using the Shapiro-Wilk test.

The results of research and their discussion: Arterial pressure in intact males were: systolic (SBP) – over 139.0±3.5 mm Hg, diastolic (DBP) – 105,0±2.9 mm Hg and HR - 420,0±9.0.The introduction of the blocker of NO-synthase, L-NAME, resulted in severe arterial hypertension (AH) (SBP – 190,3±6,7, DBP – 145,0±3,9 mm Hg and HR – 428,0±11,0) (Fig.1). During the simultaneous administration of L-NAME and ademetionine (150 mg/kg) was observed decresing values of BP (SBP to 160,2±12,6, DBP to 124.3±9,0, MBP to 136,1±10,2 mm. Hg), monotherapy with taurine (260 mg/kg) – (SBP to 158,2±5,7, DBP 122,9±5,5, MBP 134,3±5.3 mm. Hg) and with combination of ademetionine and taurine with L-NAME induced deficiency of nitric oxide indicators was reduced to the following nubers – SBP to 146,5±3,6, DBP to 126,4±3,0, MBP to 133,6±3.2 mm. Hg.Bolus intravenous injection of AH within 3-5 secondsdecreased a blood pressure, reaching a peak in the intact animals for the SBP of 84.3±1,6 for DBP of 38.7±2.8 and for mean arterial pressure (MBP) 55,8±1.7 mm. Hgwhile during the first 2-3 seconds developed sudden bradycardia up to 130-150 beats per minute. Restoring of the BP happened in average 42,2±0,8 seconds after normalization of heart rate. EIV was also characterized by a decreasingof the SBP to 87,0±2,8, DBP to 42.1±4,4, MBP to 55,7±3.5 mm Hg followed by full recovery within average 45,1±1,0 sec.Blockade of NO-synthase with the help daily during the 7-day intraperitoneal injection of L-NAME at a dose of 25 mg/kg led to a smaller decline in BP after the introduction of AH (SBP to 110,6±5,2, DBP to 82,8±6,6, MBP to 92.1±6,1 mm Hg) and NP (SBP to 88,7±4,7, DBP to 50,8,8±4,2, MBP to 63,4±4,1 mm Hg) compared to the intact group. (table1).

Tabel 1.

Dynamics of blood pressure parameters and heart rate in the simulation of deficiency of nitric oxide and correction of endothelial dysfunction

Animal group	Functional tests	DBP, mm Hg	DBP, mm Hg	HR, per minute
Intact	Source	139,0±3,5	105,0±2,9	420,0±9,0
	AH	84,3±1,6	38,7±2,8	-
	NP	87,0±2,8	42,1±4,4	-
L-NAME (25 mg/kg)	Source	190,3±6,7	145,0±3,9	428,0±11,0
	AH	110,6±5,2	82,8±6,6	
	NP	88,7±4,7	50,8±4,2	
L-NAME (25 mg/kg) + ademetionine (150 mg/kg)	Source	160,2±12,6*	124,3±9,0	320,5±15,6
	AH	94,5±5,6	66,0±6,5	
	NP	83,5±6,5	42,3±2,1	
L-NAME (25 mg/kg) + taurine (260 mg/kg)	Source	158,2±5,7	122,9±5,5	317,5±26,2
	AH	113,8±1,8*	67,3±2,4	
	NP	111,6±3,4*	46,2±1,8	
L-NAME (25 mg/kg)+ ademetionine (150 mg/kg)+ taurine (260 mg/kg)	Source	146,5±3,6	126,4±3,0	316,0±20,5
	AH	88,8±1,9	66,5±2,4	
	NP	71,0±3,8	46,0±2,1	

p<0,05in comparison with the groupL-NAME
*p<0,05in comparison with the group L-NAMEademetionine+ taurine

It was also noted potentiation of decrease in indicators of arterial pressure in response to the introduction of AH.The difference in EDV and EIV in the intact animals and animals with the introduction of the inhibitor of NO-synthase L-NAME leads to the determination of the coefficient of endothelial dysfunction (CED) as the ratio of the area of the triangle above the trend of the reduction BP reaction in response to the introduction of AH.

The CED is difference between intact group and the group treated with L-NAME in 5 times respectively of 1.2 and 5.2 in intact animals treated with L-NAME. (PokrovskayaT.G. monography).

Thus, the obtained results allow us to conclude the activation of the correction of endothelial dysfunction with ademetionine in combined application with taurine (table 2).

Table 2

Dynamics of indicators that reflect the correction of endothelial dysfunction on the background of the introduction of L-NAME with ademetionine, taurine and their combination.

Animalgroup	Functionaltests	The increase in the incidence of vascular response at Srad (mm Hg)	Timevascularreactions	Timevascularreactions	CED (the area ratio of vascular responses AH to NP)
L-NAME + адеметионин	АХ	60,8±5,2	27,4±3,7	803,5±97,0	4,6±0,5
	НП	91±8	76,8±5,1	3505,2±366,1	
L-NAME + таурин	АХ	51,9±4,5	19,7±2,1	519,2±67,9	4,3±0,7
	НП	67±4	56,1±5,1	1905,6±206,0	
L-NAME + адеметионин + таурин	АХ	59,2±2,0	41,3±7,1	1139,2±136,2	3,2±0,4*
	НП	78±4	83,3±2,4	3275,1±232,4	

*p<0,05 in comparison with the group L-NAMEademetionine+ taurine

Thus, the results demonstrate that investigational drugs have not only antihypertensive effect but also contribute to the reduction of the coefficient of endothelial dysfunction in comparison with group L-NAME (Fig.1).

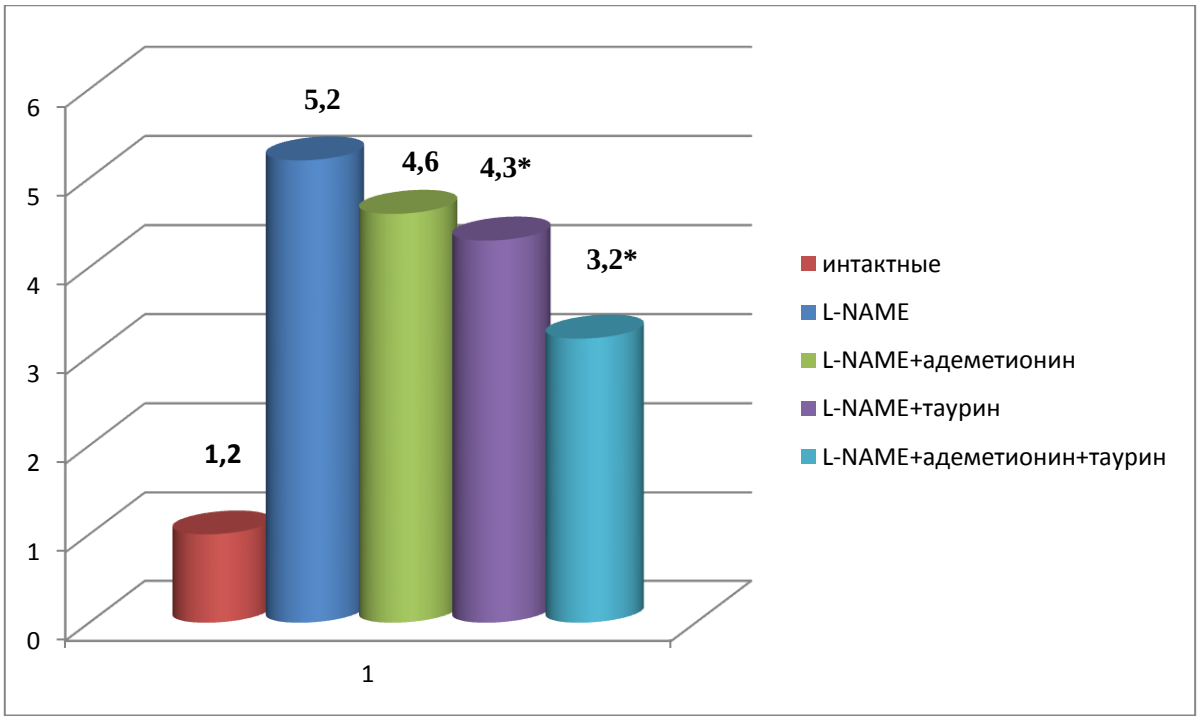

Figure 1. The values of the coefficient of endothelial dysfunction in correction with ademetionine (150 mg/kg/day), taurine (260 mg/kg/day) and their combination in comparison with the intact group and the group in modeling L-NAME (25 mg/kg intraperitoneally once daily for 7 days) induced NO deficit, *- p<0.05 compared with group L-NAME.

Conclusion: On models L-NAME-induced deficit of nitrogen oxide are realized antihypertensive, antioxidant andendothelioprotectiveproperties of taurine and ademetionine, as according to the literature, inhibition of nitric oxide production in the application of L-NAME was accompanied by a significant increase in the spontaneous production of superoxide anion radical, hypertension and increase of coefficient of endothelial dysfunction [10, 20]. Hyperproduction of superoxide radical and its derivatives – oxygen radicals – is a mechanism for development of oxidative stress (OS), in which the suppression of the antioxidant defense system and increased formation of oxidation products, which arethe factors for the formation of endothelial dysfunction [21, 22, 23].In the study it was found that the combined using of ademetionine in the dose 150mg/kg/day and taurine at a dose of 260 mg/kg/day to provides more endothelioprotective effect on models L-NAME-induced deficit of nitrogen oxide in comparison with monotherapy, which was reflected in the decrease of coefficient of endothelial dysfunction (CED) [24] and the BP values to a level close to the level of intact animals.Therefore, monotherapy endothelial dysfunction with ademetionine and taurine is regarded as insufficient, and leads to further search for more effective ways of pharmacotherapy, one of which is the combined application of taurine and ademetionine. From our point of view, the combination of ademetionine and taurineis warranted to examine itsendothelio - and cardioprotective effect.

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