and chromatography on Protein A-Sepharose can be used in clinical practice without any significant loss of sensitivity of the determination, because of the "moldness" of these methods, which cause little disturbance of the organization of the active centers of the immunoglobulins.

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# SUPEROXIDE SCAVENGING ACTIVITY AND TRANSFERRIN–CERULOPLASMIN ANTIOXIDANT SYSTEM IN RAT SERUM DURING CHRONIC EMOTIONAL-PAINFUL STRESS AND DIMETHYL SULFOXIDE TREATMENT


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The state of the antioxidant systems of the blood serum reflects the general antioxidant status of the body and changes during exposure to external environmental factors, thereby ensuring the resistance of the body to such exposure. It was demonstrated in [1] that the ratio between the serum levels of ceruloplasmin (Cp) and transferrin (Tr) is an indicator of serum antioxidant activity and reflects the resistance of rabbits to hypercholesterolemia. It has recently been shown that blood serum possesses superoxide-scavenging activity (SSA), and this evidently also makes an essential contribution to the antioxidant potential of the blood serum [4]. The aim of the present investigation was to study the state of these systems of antiradical defense of the blood in rats exposed to chronic emotional-painful stress (EPS) and treated with dimethyl sulfoxide (DMSO), a scavenger of hydroxyl radicals, in order to correct EPS-induced disturbances.

## EXPERIMENTAL METHOD

Altogether 30 noninbred male albino rats weighing 200-250 g were used. EPS was produced by combined action of electrodermal stimulation and white noise, as described previously [2], for 3 weeks. The Cp and Tr levels were measured by EPR-spectrometry, under the conditions described in [1]. The SSA of whole blood serum was determined in a system containing adrenalin under auto-oxidation conditions [3] or in a phenazine metasulfate–NADH–nitroblue tetrazolium system [5]. To determine nonprotein SSA, the blood serum proteins were completely precipitated with TCA, the serum was neutralized with KOH, and SSA was determined in the protein-free supernatant as described above.


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TABLE 1. Serum Cp, Tr, and SSA Levels in Rats Subjected to EPS and Treated with DMSO

| Parameter | Control | EPS | Control + DMSO | EPS + DMSO |
|---|---|---|---|---|
| Cp, conventional | 2,56±0,27 | 2,54±0,28 | 2,43±0,18 | 2,51±0,28 |
| Tr, conventional units | 7,68±0,45 | 8,69±0,97 | 9,07±0,49* | 9,71±1,08*** |
| Cp/Tr | 0,34±0,04 | 0,37±0,05 | 0,27±0,02** | 0,28±0,03** |
| SSA of whole serum, U/ml | 5,5±0,2 | 5,0±0,4 | 6,5±0,5** | 8,0±0,9** |
| SSA of protein-free supernatant, U/ml | 9,6±0,1 | 8,3±0,1 | 11,7±0,4** | 13,1±0,3** |

Legend. Levels of significance of differences: *p < 0.02, **p < 0.01 compared with control; ***p < 0.02, ****p < 0.01 compared with EPS group.

## EXPERIMENTAL RESULTS

Animals exposed to chronic EPS showed disturbances of the cardiovascular, respiratory, and autonomic nervous systems similar to those arising in neurosis-like states [2]. Injection of DMSO into the animals in a dose of 1 g/kg before each EPS session prevented the development of disturbances of visceral functions (Table 1).

The experiments showed that EPS caused no significant changes in the antioxidant system. There was only a tendency for the Tr level to rise (by 13%) and for the Cp/Tr ratio to fall (by 9%). SSA of whole serum and of the protein-free supernatant likewise showed a tendency to fall (by 9 and 14%, respectively).

SSA of the serum increased by 65-80% after precipitation of the protein. The serum SSA correlated with the "nonprotein" SSA (r = 0.76).

Injection of DMSO into the control rats led to a significant rise of the Tr level by 18%, a fall of the Cp/Tr ratio by 21%, and an increase in SSA in whole blood serum and in the protein-free supernatant by 18 and 22%, respectively. In the animals subjected to EPS, injection of DMSO caused the Tr level to rise and the Cp/Tr ratio to fall by 12 and 24%, respectively, just as in the control rats. SSA of whole blood serum and protein-free SSA increased significantly more than in the control animals — by 60 and 59%, respectively.

Analysis of the results obtained in control rats and rats subjected to EPS showed correlation between the Tr level and SSA of whole blood serum (r = 0.65). However, a separate experiment shows that addition of Tr to the blood serum, accompanied by an increase in the degree of its iron saturation, did not lead to an increase in SSA, but in the presence of a high degree of saturation it inhibited SSA. This is evidence that SSA is not connected with the presence of Tr in the blood serum.

Significant correlations were found also between the Cp/Tr ratio and SSA of whole blood serum (r = −0.59) and also between the protein-free SSA and Tr level (r = 0.63) and Cp/Tr ratio (r = 0.56). No correlation was found between the Cp level and SSA. These data indicate that the Cp/Tr system and the system possessing SSA are two independent systems of the serum.

This investigation thus demonstrated interaction in vivo between two systems of antiradical defense of the blood serum during exposure of the animal to EPS and treatment with the hydroxyl radical scavenger DMSO.

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