ROLE OF CERULOPLASMIN, TRANSFERRIN, AND LIPID PEROXIDATION IN BACTERIAL INFECTIONS OF THE CNS

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Worsening of the state of patients with bacterial infections of the CNS is associated with involvement of individual organs and systems, which is accompanied by the appearance of enzymes in the blood [3]. The appearance of raised blood enzyme levels in pathology is linked with disturbance of the barrier function of the membranes. A fundamental mechanism of cytolysis is lipid peroxidation (LPO). The level of LPO in the body is controlled by anti- and pro-oxidant systems, of which ceruloplasmin and transferrin are representatives respectively [1].

EXPERIMENTAL METHODS

Blood from 40 patients with bacterial diseases of the CNS of meningococcal and pneumococcal etiology was studied on the 1st-3rd, 5th, 7th, 10th, 15th, and 20th days of the disease. The patients' ages ranged from 19 to 56 years. The level of aspartate transaminase (AST) activity was determined on an "Olli-3000" biochemical analyzer (Kohe, Finland) by a kinetic method based on the change in extinction at 340 nm, using kits from Kohe and expressed in units per liter of blood. The concentration of LPO products was determined as the level of malonic dialdehyde (MDA), expressed in micromoles/liter, the molar coefficient of extinction being taken to be $1.56 \cdot 10^5$ $mole^{-1} \cdot cm^{-1}$.

To measure the transferrin and ceruloplasmin concentrations the method of EPR-spectroscopy was used. Preparations for EPR-spectroscopy were made in tablet form, by freezing blood serum in liquid nitrogen. The tablet was placed in a Dewar flask with liquid nitrogen, which was introduced into the resonator of a "Varian E-4" spectrometer. The conditions of measurement were: frequency of the klystron generator 9.03 MHz, power 10 mW, amplitude of modulation 6.3 G, field scanning speed 250 G/min, time constant of the instrument 3.0 sec. On the spectra thus obtained the signal with $g = 2.05$ belonged to ceruloplasmin, that with $g = 4.3$ to transferrin. The amplitude of the EPR signals was next measured, and taken to be proportional to the concentration of paramagnetic centers.

Quantitative determination of the plasma protein composition of patients with bacterial infections of the CNS was carried out by the method of two-dimensional gel electrophoresis as described in [4].

The results were subjected to standard statistical analysis with calculation of the arithmetic mean, the standard deviation, and the significance of changes by Student's test on an Olivetti computer (Italy).

EXPERIMENTAL RESULTS

The study of the time course of the serum ceruloplasmin and transferrin concentrations of the patients revealed curves of different character (Fig. 1). The ceruloplasmin level

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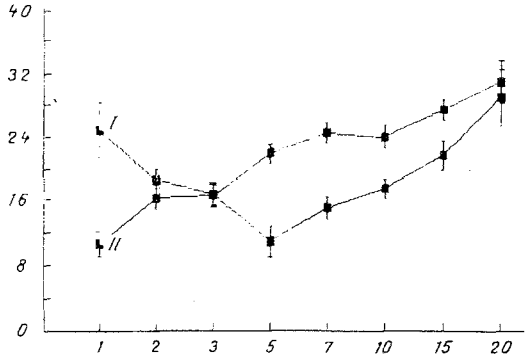

Fig. 1

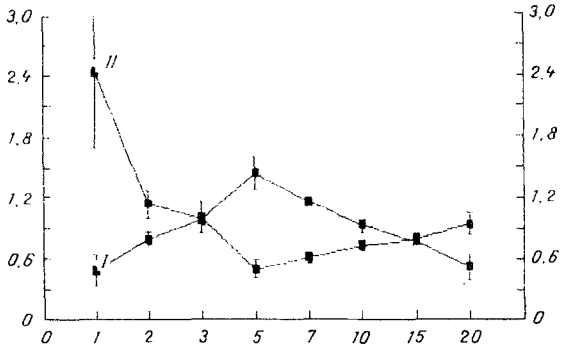

Fig. 2

Fig. 1. Changes in plasma ceruloplasmin (I) and transferrin (II) concentrations in course of bacterial infection of the CNS. Abscissa) time (in days); ordinate) concentration (in r.u.).

Fig. 2. Time course of relative concentrations of ceruloplasmin and transferrin (II) and levels of LPO products (I) during disease. Abscissa) time (in days); ordinate) on right – ratio (in r.u.); on left – MDA level (in μmoles/liter).

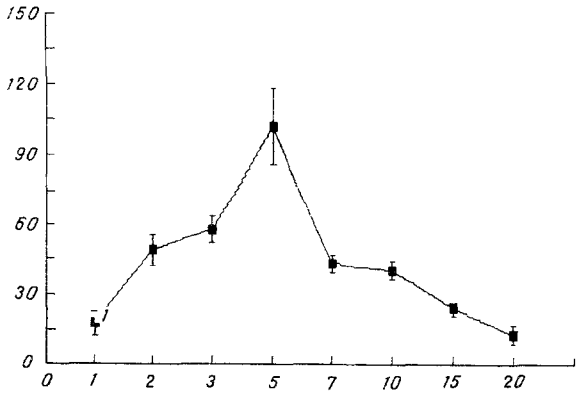

Fig. 3. Level of AST activity during course of disease. Abscissa) time (in days); ordinate) activity (in U/liter).

on the 1st day of the disease was 24.8 ± 3.3 relative units (r.u.), it fell gradually by the 5th day to 10.9 ± 1.8 r.u., and then rose to its peak values of 28.5 ± 3.5 r.u. by the 20th day. The transferrin concentration rose gradually from the 1st through the 20th day from 10.6 ± 1.6 to 30.3 ± 2.5 r.u.

Considering that LPO depends on the concentrations of ceruloplasmin and transferrin, as representatives of the anti- and pro-oxidant systems, the ratio between them was calculated during the course of the disease (Fig. 2). On the 1st day of the disease the ceruloplasmin/transferrin ratio was highest (2.43 ± 0.74), but later it fell sharply toward the 5th day – to 0.50 ± 0.09 – and then rose by the 20th day to 0.94 ± 0.11. This character of the change in the ratio was opposite to the time course of the change in MDA. The MDA level on the 1st day was 0.49 ± 0.16 μmole/liter, rising by the 5th day to 1.45 ± 0.13 μmole/liter, and by the 20th day it fell to 0.51 ± 0.11 μmole/liter. The coefficient of correlation between the change in the MDA level and the ceruloplasmin/transferrin ratio was −0.69. Elevation of the MDA level toward the middle of the disease may perhaps be associated with a considerable fall in the ceruloplasmin/transferrin ratio during the same period. This can be explained by the fact that the transferrin level (the pro-oxidant) was higher than the ceruloplasmin level.

The character of the time course of the ceruloplasmin/transferrin ratio was mainly determined by the time course of the ceruloplasmin concentration (Figs. 1 and 2). The coefficient of correlation between the MDA and ceruloplasmin concentrations in the blood serum was −0.96, whereas the coefficient of correlation between MDA and transferrin was 0.10. On the 5th day of the disease the maximal MDA level corresponded to the minimal value of the ceruloplasmin/transferrin ratio. The subsequent fall of the MDA level coincided with an increase in this

coefficient. This change in the ceruloplasmin/transferrin ratio was determined by the time course of the ceruloplasmin concentrations, for the transferrin concentration rose smoothly from the 1st through the 20th days. The rise of the MDA level toward the 5th-7th days coincided with a sharp decline in the ceruloplasmin concentration, possible evidence of intensification of LPO and of depression of activity of the antioxidant system. After the middle of the disease, and up to the end of the 20th day, the time course was opposite in nature. It will be clear from Fig. 1 that this was due to the more rapid rise of the ceruloplasmin than of the transferrin concentration. The LPO level in cells and tissues of the body is mainly determined by the free Fe^{++} level and it may be regulated by systems responsible for oxidation and reduction of iron. Fe^{+++} is carried in the composition of transferrin to various tissues by the blood flow [1]. The rise of the transferrin level from the beginning until the end of the disease (Fig. 1) is evidently linked with the need to transport Fe^{+++}. The increase in the iron concentration may perhaps be due to massive destruction of erythrocytes throughout the course of the disease [3]. Periodic observations of the protein composition of the blood plasma revealed a large quantity of haptoglobin toward the end of the disease, i.e., toward the time of maximal rise of the transferrin level. This indicates the need to maintain globin after the cytolysis which took place at the height of the disease. The reason for the lower transferrin level at the beginning of the disease may be competition between pathogenic bacteria and the host organism for iron [2], in the course of which iron-chelating proteins (siderophores) remove iron from transferrin with 30% saturation.

Elevation of the MDA level against the background of a sharp decline in the ceruloplasmin/transferrin ratio in patients with bacterial diseases of the CNS coincides with times of a raised AST level (Fig. 3). This may perhaps confirm the presence of massive cytolysis against the background of LPO activation and may correspond clinically to the height of the disease.

LITERATURE CITED

1. Yu. A. Vladimirov, "The role of disturbance of the barrier and matrix functions of the lipid layer of biological membranes in pathology," Degree Oration [in Russian], Moscow (1985).
2. G. Dick (ed.), Immunological Aspects of Infectious Diseases, Univ. Park, Baltimore (1979).
3. V. I. Pokrovskii, L. A. Favorova, and N. N. Kostyukova, Meningococcal Infection [in Russian], Moscow (1976).
4. N. P. O'Farell, J. Biol. Chem., 250, No. 10, 4007 (1975).