

## EXPERIMENTAL METHODS FOR CLINICAL PRACTICE

# Correlation Between the Parameters of Free-radical Lipid Peroxidation and Antioxidant System in Children Living in the North

G. I. Bisharova, L. I. Kolesnikova, and V. V. Malyshev

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 126, No. 9, pp. 342-344, September, 1998  
Original article submitted November 10, 1997

A comparative study of the parameters of free-radical lipid peroxidation and antioxidant system was performed in children living in the North for various time periods. Intense lipid peroxidation was shown to be the key factor in the pathogenesis of several diseases caused by disturbances in the cellular membrane. Decreased resistance of red blood cells to peroxidative hemolysis is a phenomenon characterizing the adaptation-violating processes.

**Key Words:** correlation; lipid peroxidation; antioxidant activity; children; North

Children are more sensitive to hazardous factors affecting their health because the mechanisms of autoregulation are not fully developed [4,8]. Long-term effects of extreme factors of the northern latitudes induce systemic reconstructions of oxygen conditions of cells to provide a new bioenergetic level and intensify of lipid metabolism and free-radical lipid peroxidation [6]. The biochemical parameters of lipid metabolism, oxidative status (the intensity of lipid peroxidation, LPO), and universal antioxidant system (antioxidant activity, AOA) reflect the intensity of free-radical processes in individual organs and systems as well as in the whole body. Intensification of these processes contributes to the development (pathogenesis) of many diseases. On the other hand, this intensification shows a type of adaptive reactions to various stress stimuli.

This work was designed to study the interrelations between the parameters of LPO and AOA in blood plasma and erythrocytes obtained from children living in the North.

Chita Affiliate of the Institute of Pediatrics and Human Reproduction, Eastern Siberian Scientific Center, Russian Academy of Medical Sciences; Institute of Pediatrics, Siberian Division of the Russian Academy of Medical Sciences, Irkutsk

## MATERIALS AND METHODS

We examined 103 healthy children at the age of 7-11 years living in the North: 20 Evenki, group 1; 20 Russian permanent residents, group 2; 20 Russian nonresident children, group 3; and 43 children of the same age living in the city of Chita, control group.

The contents of LPO products was measured by extraction and spectrophotometry in heptane and isopropanol phases of lipid extract [3]. The contents of total lipids and total phospholipids and the protein concentration were determined by combined methods using special kits.

The concentration of thiobarbituric acid-reactive substances (TBARS) was measured as described [1]. The content of LPO products extracted with heptane and isopropanol was expressed in units of oxidative index. This index was calculated in the corresponding phases of the lipid extract as the ratio of optical densities of  $E_{232}/E_{220}$  (for conjugated dienes), and the ratio of optical densities of  $E_{278}/E_{220}$  (for ketodienes and conjugated trienes). Peroxidative resistance of erythrocytes was assessed by the method [5]. AOA was determined by analyzing the ability of plasma or erythrocytes to inhibit per-

oxidation of biogenic lipids *in vitro* [2,7]. Mathematical analysis was performed by the method of variational statistics; the differences were evaluated using the Student's *t* test. Correlation dependences (*r*) were estimated by the Pearson test.

## RESULTS

While analyzing the correlations between the parameters studied, we did not consider logical dependences which were known in advance (for example, conjugated dienes, the  $E_{232}/E_{220}$  ratio; and ketodienes and conjugated trienes, the  $E_{278}/E_{220}$  ratio).

Analysis of the coefficients of correlation in the control group revealed quite regular interrelations between levels of primary (conjugated dienes) and secondary (ketodienes and conjugated trienes) products of LPO in the isopropanol fraction of lipid extract ( $r=0.91$ ,  $p<0.001$ ); conjugated dienes in this phase and the  $E_{278}/E_{220}$  ratio ( $r=0.55$ ,  $p<0.05$ ); and ketodienes, conjugated trienes, and the  $E_{232}/E_{220}$  ratio ( $r=0.61$ ,  $p<0.02$ ). The relationship between the content of erythrocytic TBARS and the activity of catalase was sufficiently strong ( $r=0.90$ ,  $p<0.001$ ). Under physiological conditions, the equilibrium between LPO and antioxidants is a constant [9]. Thus, this dependence is well substantiated.

Similar particularities of the interrelations between the components studied were revealed in permanent residents (Table 1). In contrast to the Russians displaying three significant correlation dependences, the Evenki had greater number of dependences. Two of these dependences (between plasma contents of total lipids and TBARS; and between the  $E_{232}/E_{220}$  ratio and secondary LPO intermediates) are of interest. The correlations can be interpreted as follows: the first correlation suggests

that the level of LPO products in the Evenki tends to decrease or remains unchanged (in spite of the increase in the content of substrates for free-radical reactions). The second correlation implies that the processes of radical oxidation of lipids are strongly interrelated, since an increase in the level of secondary products induces a decrease in the concentration of primary products of these reactions (and vice versa).

The greatest number (10) of correlations was shown in the third group. Intermediate positive correlations between the level of conjugated dienes in the isopropanol fraction and erythrocytic TBARS ( $r=0.52$ ,  $p<0.05$ ), plasma catalase ( $r=0.64$ ,  $p<0.02$ ), and erythrocyte catalase ( $r=0.52$ ,  $p<0.05$ ) were observed. Similar dependencies between the  $E_{232}/E_{220}$  ratio and the content of intermediate products of LPO, the percent of erythrocytes hemolyzed in the reaction of peroxidative resistance of erythrocytes and the content of TBARS in erythrocytes, and activities of erythrocytic catalase and superoxide dismutase were revealed. Strong positive correlations between the level of erythrocytic TBARS and activities of catalase in erythrocytes ( $r=0.94$ ,  $p<0.001$ ) and blood plasma ( $r=0.70$ ,  $p<0.01$ ) were observed. The latter correlation depended on the rate of  $H_2O_2$  detoxification in erythrocytes ( $r=0.81$ ,  $p<0.01$ ). A negative correlation between the content of total lipids and the activity of plasma catalase seemed to be unusual ( $r=-0.50$ ,  $p<0.05$ ). It was possible to reveal some basic principles of the development of the syndrome of lipid hyperperoxidation inducing destruction of membranes and hemolysis in nonresident children.

Initiation of LPO induces the increase in the contents of primary, secondary, and intermediate products of LPO. This activates the factors of the antioxidant system which are then depleted. The

TABLE 1. Coefficients of Correlation of LPO and AOA Indices in Children Living in Northern Regions of Zabaikal'e

Parameters correlated	Russian permanent residents	Evenki
Conjugated dienes, ketodienes, and conjugated trienes	0.94***	0.76**
Conjugated dienes and $E_{278}/E_{220}$	0.57*	—
$E_{278}/E_{220}$ and plasma catalase	0.56*	—
Total lipids and TBARS of plasma	—	-0.59*
Conjugated dienes and TBARS of plasma	—	0.73**
$E_{232}/E_{220}$ , ketodienes, and conjugated trienes	—	-0.86**
Ketodienes, conjugated trienes, and TBARS of plasma	—	0.70*
Ketodienes, conjugated trienes, and erythrocytic catalase	—	0.50*
TBARS of plasma and plasma catalase	—	0.55*
TBARS of plasma and erythrocytic catalase	—	0.50*

Note. \*\* $p<0.05$ , \*\*\* $p<0.01$ , \*\*\*\* $p<0.001$ , \* $p<0.2$  significant differences; — insignificant differences.

depletion contributes to the development of structural and functional alterations in biological membranes, for example, a decrease in the resistance of red blood cells to peroxidative hemolysis.

The data obtained provide additional insights into the mechanisms of the general adaptation syndrome involving the change in the type of biochemical reactions and the decrease in the resistance of homeostatic systems.

## REFERENCES

1. L. I. Andreeva, L. A. Kozhemyakin, and A. A. Kishkun, *Lab. Delo*, No. 11, 41-43 (1988).
  2. V. P. Verbelovich and L. M. Podgornaya, *Ibid.*, No. 2, 17-20 (1987).
  3. I. A. Volchegorskii, A. G. Nalimov, B. G. Yarovinskii, and R. I. Livshits, *Vopr. Med. Khimii*, No. 1, 127-131 (1989).
  4. N. A. Dikaya, T. V. Gordeeva, N. I. Katulina, *et al.*, *Vopr. Okhr. Mat.*, No. 10, 66-67 (1982).
  5. *Study of Indices of Lipid Metabolism and Lipid Peroxidation. Method. Recommend.* [in Russian], G. A. Yarovaya (Ed.), Moscow (1987).
  6. V. P. Kaznacheev, *Actual Aspects of Adaptation* [in Russian], Novosibirsk (1980).
  7. M. A. Korolyuk, L. I. Ivanova, I. G. Maiorova, and V. E. Tokarev, *Lab. Delo*, No. 1, 16-19 (1988).
  8. V. I. Krylov, *Pediatrics*, No. 4, 57-58 (1980).
  9. M. Comporti, *Mol. Aspects Med.*, **14**, No. 3, 199-207 (1993).
-