# LIPID PEROXIDATION IN THE AORTA OF NORMOTENSIVE AND SPONTANEOUSLY HYPERTENSIVE DIABETIC RATS

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UDC 616.379-008.64-092.9-06:616.12-008.331.1]-07:616.132-008.939.15-39

**KEY WORDS:** lipid peroxidation; aorta; rats; hypertension; diabetes.

The process of lipid peroxidation (LPO) is widely distributed and takes place to some degree in all cells, although not very rapidly. In diabetes (D) particular importance is attached to disturbances of lipid metabolism. According to one hypothesis of the onset of D, involvement of blood vessels (observed whenever tolerance to glucose is disturbed) arises as a result of hereditary traits of metabolism in the vascular wall and, in particular, accumulation of low- and very low-density lipoproteins [3]. A definite role in this situation may perhaps be played by autooxidation of polyunsaturated fatty acids of cell membranes. It has been shown, for instance, that the plasma LPO level is raised in diabetics, and that concentrations of lipid peroxides were higher still on the appearance of retinopathies [13].

In the experiments described below concentrations of conjugated dienes (CD) and malonic dialdehyde (MDA) were determined and the percentage of formation of Schiff's bases in the aortic tissue of normotensive (Wistar–Kyoto strain, MR) and spontaneously hypertensive (Okamoto–Aoki strain, SHR) rats was established during the development of streptozotocin-induced diabetes in the animals.

## EXPERIMENTAL METHOD

Insulin-dependent streptozotocin-induced diabetes was elicited by a single intraperitoneal injection of streptozotocin, made up when required at the rate of 60 mg/kg body weight in 0.5 ml of citrate buffer, pH 4.2 [11]. This dose of streptozotocin caused a permanent rise of the blood glucose level, without any general toxic action on the animals. Not all the animals developed D (rather more than 50%). In the group developing D they were rats whose blood glucose level 1 week after injection of streptozotocin was not less than 250 mg/dl. The blood pressure was measured in the caudal artery by means of an electrosphygmomanometer (Narco BioSystems, USA). The experiment lasted 6 weeks. The rats were killed under superficial ether anesthesla, the aorta was isolated and washed with ice-cold 0.85% NaCl solution to remove blood, dried with filter paper, and homogenized in a glass homogenizer on ice. For every 10 mg tissue 1 ml of phosphate buffer, pH 7.6, was taken. MDA and CD in the homogenate were determined by the method in [4], Schiff's bases as in [7], and the total lipid content with the aid of ready-made kits from Reanal. The results were subjected to statistical analysis by Student's t test. The results were subjected to correlation analysis with calculation of the coefficient of correlation "r."

## EXPERIMENTAL RESULTS

The experiments showed that development of D leads to an increase in the total lipid content per milligram tissue in the aorta by 109.4% in NR and by 38.4% in SHR (Table 1). Correlation was absent in the control NR between the blood glucose level and the total lipid content in the aortic tissue. Meanwhile, in the control SHR, positive correlation was found from the beginning between these parameters (r = +0.545). The development of D

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(Presented by Academician of the Academy of Medical Sciences M. Ya. Studenikin.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 113, No. 5, pp. 495-496, May, 1992. Original article submitted February 28, 1991.


0007-4888/92/0005-0666$12.50 

TABLE 1. Content of Lipid Peroxidation Products in Aorta of Normotensive and Spontaneously Hypertensive Diabetic Rats (M ± m)

| Parameter | Rats | | | |
|---|---|---|---|---|
| | normotensive | | spontaneously hypertensive | |
| | control | diabetes | control | diabetes |
| Content of total lipids/mg tissue | 0,0085±0,0009 (n=14) | 0,0169±0,003** (n=8) | 0,0091±0,0009 (n=17) | 0,0124±0,002 (n=11) |
| CD, μmoles/mg total lipids | 0,48±0,082 (n=15) | 0,73±0,7* (n=8) | 1,07±0,22** (n=11) | 0,87±0,37 (n=7) |
| MDA, μmoles/mg total lipids | 0,36±0,036 (n=12) | 0,66±0,14* (n=7) | 0,63±0,12* (n=13) | 0,64±0,2 (n=7) |
| Percent of Schiff bases/mg total lipids | 18,1±1,5 (n=13) | 22,6±3,4 (n=8) | 36,8±3,9** (n=13) | 30,7±4,3 (n=10) |

**Legend.** *$p < 0.05$ Compared with control; **$p < 0.05$ for comparison of NR and SHB.

and elevation of the blood pressure in MR to 145.5 ± 5 mm Hg led to the appearance of definite correlation between the blood glucose level and the total lipid content in the aorta ($r = +0.23$). In SHR, despite their generally severe state caused by the development of D, no further rise of blood pressure was observed above 163.8 ± 12.4 mm Hg; no correlation was found between the blood glucose level and the total lipid content in the aorta.

There is evidence that after intravenous injection of glucose changes of diabetic type in the sugar curve are observed in patients with essential hypertension [6], and that in members of hypertensive families an increase in the total lipid content has been found in erythrocyte membranes on account of the cholesterol friction, and this may increase the microviscosity of the membranes [1]. This is in agreement with our data showing correlation between the total lipid level in the aortic tissues and the plasma glucose level in control SHR and NR with developed diabetes. Meanwhile, the development of insulin-dependent diabetes, which includes streptozotocin diabetes of rats, is accompanied by increased flowability of the membranes [12], while at the same time, the total lipid content in the aorta showed a smaller increase in SHR than in NR, i.e., SHR were already adapted to some extent to changes induced by the development of diabetes compared with NR.

Judging by the formation of CD, MDA, and Schiff bases, LPO processes in the control SHR took place much more actively than in NR. For instance, CD formation in SHR was increased by 2.2 times ($p < 0.001$) MDA formation by 1.75 times ($p < 0.05$), and the percentage formation of Schiff bases was doubled ($p < 0.001$) compared with the corresponding SHR group. The discovery of conjugated dienes in biological membranes is regarded as a sensitive test for the appearance of hydroperoxides, and one of the most important functions of the latter is regulation of the prostacycline:thromboxane ratio. In the normal aorta this ratio is 74:1, but with intensification of LPO it changes to 10:1 [9, 14]. Low levels of peroxides, it is claimed, stimulate prostaglandin synthesis, whereas high levels block it [10]. It has also been suggested [5] that activation of LPO, which we observed in the control SHR, may be the chief cause of disturbance of the functions of the Ca-pump of the sarcoplasmic reticulum, which is usually observed in long-lasting arterial hypertension. Negative correlation was found in the control NR between the CD content and the percentage formation of Schiff bases in the aortic tissues ($r = -0.48$); no such correlation was observed in SHR of the corresponding group.

The development of diabetes in spontaneously hypertensive and normotensive animals was accompanied by opposite changes in the formation of LPO products. In NR, CD formation was increased by 1.5 times ($p < 0.05$) and MDA formation by 1.7 times ($p < 0.05$); the formation of Schiff bases was increased, but not significantly. In diabetic SHR there was actually a small decrease in CD and Schiff base formation, but this decrease likewise was not statistically significant.

In the group of NR with developed diabetes, definite correlation appeared between the blood sugar level and concentrations of: MDA ($r = +0.68$, $p < 0.01$), CD ($r = +0.76$, $p < 0.05$), and percentage of Schiff bases ($r = +0.70$, $p < 0.05$). Correlation also was established between CD and the percentage of Schiff bases ($r = +0.79$, $p < 0.05$) and between the MDA and CD levels ($r = +0.77$, $p < 0.05$). No correlation was found between the formation of MDA and of Schiff bases. In diabetic SHR, by contrast with the corresponding group of MR, no correlation was

found between MDA, CD, and Schiff bases, but an equal degree of correlation was found between the blood glucose level and concentrations of MDA and Schiff bases ($r = -0.64$, $p < 0.05$). In SHR with an extreme degree of adaptation to a permanently raised blood pressure (hereditarily determined) development of diabetes leads to failure of the adaptive reactions [2]; this may perhaps account for the great sensitivity of SHR to streptozotocin [8] and to the more severe course of diabetes as a whole.

The development of diabetes in normotensive and spontaneously hypertensive rats is thus accompanied by opposite changes in the intensity of LPO, possible evidence of failure of adaptation of spontaneously hypertensive animals during the development of diabetes.

## LITERATURE CITED


1. Yu. M. Bala and G. I. Furmenko, Calcium Metabolism in the Physiology and Pathology of the Cardiovascular System [in Russian], Part 1, Tomsk (1988), pp, 29-30.
2. F. Z. Meerson, Adaptation, Stress, Prophylaxis [in Russian], Moscow (1981).
3. I. D. Saltykov, Arkh. Patol., **46**, No. 8, 75 (1984)
4. I. D. Stal'naya, Modern Methods in Biochemistry [in Russian], Moscow (1977), pp. 63-68.
5. I. L. Tverdislova and V. B. Ritov, Byull. Éksp. Biol. Med., **103**, No. 4, 415 (1987).
6. R. Baumann, et al., Dtsch. Gesund.-Wes., **26**, 525 (1971).
7. W. R. Bidlack and A. L. Tappel, Lipids, **8**, No. 4, 203 (1973).
8. M. E. Cooper et al., Clin. Exp. Pharmacol. Physiol., **3**, No. 9, 655 (1986),
9. R. Maddipati and L. J. Marnett, J. Biol. Chem., **262**, 17398 (1987).
10. S. Murota, Prostagland Leukotr. Med., **25**, No. 2/3, 123 (1986).
11. J. R. Sedor, J. Lab. Clin. Med., **108**, No, 2, 521 (1986).
12. I. Testa et al., J. Clin. Endocr., **67**, No. 6, 1129 (1988).
13. N. Uzel et al., Horm. Metab. Res., **19**, No. 2, 89 (1987).
14. J. Wang et al., Exp. Molec. Path., **48**, No. 2, 153 (1988).


# EFFECT OF ARGININE ON PROPERTIES OF ERYTHROCYTE MEMBRANES IN HYPOXIA


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UDC 615.31:547.495.9].015.4:[616.155.1-018.1:576 314]-092:612.273 2].076.9

**KEY WORDS:** hypoxia; arginine; erythrocyte membranes; lipid peroxidation; superoxide dismutase.


Potentiation of free-radical processes with an increase in the intensity of lipid peroxidation (LPO) is observed in hypoxia [2], and this, in turn, may cause many structural and metabolic changes, including changes in membrane permeability [12] and changes in activity of enzyme systems located in membranes [1]. Arginine has been shown to have a protective action in many functional states [9]. We have demonstrated the antihypoxic effect of arginine [12]. The influence of arginine on lipid metabolism has been demonstrated [3]. Its antiradical and antioxidant effects have been established by experiments in vitro [10].


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(Presented by Academician of the Academy of Medical Sciences T. T. Berezov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 113, No. 5, pp. 497-498, May, 1992. Original article submitted October 18, 1991.




0007-4888/92/0005-0668$12.50