Membrane-Stabilizing Effect of δ-Sleep-Inducing Peptide in Stress

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Exposure of rats to cold stress leads to the accumulation of conjugated dienes and Schiff's bases and decreases superoxide dismutase and catalase activities. Plasma levels of extraerythrocytic hemoglobin and iron and the total peroxidase activity increase. This indicates destabilization of the erythrocyte plasma membrane in stress. Exogenous δ-sleep-inducing peptide decreases the intensity of lipid peroxidation by increasing the activity of the antioxidant enzymes and stabilizes the structure of the erythrocyte plasma membrane.

Key Words: *δ-sleep-inducing peptide; stress; erythrocyte membranes*

The relationship between stress, health, and diseases has been extensively investigated. It was shown that 90% of diseases are related to stress [10]. δ-Sleep-inducing peptide (DSIP), an antistress and hypnagogic brain peptide modulator, is of special pharmacological interest. The use of regulatory peptides is a physiological and directed therapeutic method.

The activation of lipid peroxidation (LPO) and free-radical reactions inducing damage to biological membranes plays an important role in the stress reaction. Many pathological changes involve cell membranes and alter their molecular structure. This increases the hazardous effects of diseases [7]. Our previous studies demonstrated a membrane-stabilizing effect of DSIP in stress [2]. The mechanism of this effect is unclear. Therefore, we studied the effects of DSIP on the content of LPO products in erythrocyte membranes; the activity of the erythrocyte antioxidant enzymes, superoxide dismutase (SOD), catalase, and glucose-6-phosphate dehydrogenase (G-6-PDH), the levels of extraerythrocytic hemoglobin and iron and the total plasma peroxidase activity in rats under normal conditions and in stress.

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MATERIALS AND METHODS

Experiments were performed on adult albino rats of both sexes weighing 150-180 g. Control and treated animals were divided into 4 groups: group 1, intact rats (vivarium, food and water *ad libitum*); group 2, intact rats injected with DSIP (12 μg/100 g body weight, i. p.) and decapitated 3 days after the injection; group 3, rats subjected to a 3-day cold stress (0-4°C); and group 4, rats injected with DSIP (12 μg/100 g body weight, i. p.) immediately before the exposure to cold. The erythrocyte membranes were prepared as described [6]. Lipids were extracted from blood plasma and erythrocyte membranes [11]. The activity of LPO was determined by calculating the contents of LPO primary (conjugated dienes) and fluorescent end products (Schiff's bases) [4]. The total lipid level was measured by the phosphovanillin method [5]. The total peroxidase activity [8] and the activities of SOD [12], catalase [14], and G-6-PDH [3] were assayed. Protein concentration was estimated by a modified method of Lowry [13]. Standard kits were used for measuring hemoglobin and iron concentrations.

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Table 1. Effects of DSIP on Contents of LPO Products, Activities of Erythrocyte Antioxidant Enzymes, and Indices of the Erythrocyte Membrane Stability in Rats Subjected to Cold Stress (*M±m*, *n*=8-10).

Index	Control	Control+ DSIP	Cold stress	Cold stress+ DSIP
Erythrocytes				
Conjugated dienes, nmole/mg lipid	6.11±0.32	2.31±0.29*	10.24±0.71*	3.97±0.55*
Schiff's bases, relative units of fluorescence/mg lipid	1.55±0.12	1.04±0.14*	2.63±0.28*	2.16±0.12*
Catalase, U activity/mg hemoglobin	50.18±1.92	53.49±1.58	42.23±1.56*	53.49±1.5
SOD, U activity/mg hemoglobin	7.57±0.65	10.00±0.68*	3.81±0.39*	7.25±0.65*
G-6-PDH, nmole NADPH/mg protein/min	0.087±0.005	0.129±0.11*	0.234±0.013*	0.112±0.008*
Plasma				
Extraerythrocytic hemoglobin, mg/100 g	22.91±1.05	21.63±0.78	37.30±1.74*	22.94±1.08
Iron, μmole/l	9.10±0.95	8.90±0.72	18.60±1.27*	9.50±0.52
Total peroxidase activity, U activity/mg protein	3.75±0.14	3.68±0.11	13.18±0.27*	4.21±0.11*

Note. *Statistically significant differences (compared with the control).

RESULTS

Cold stress activated LPO processes in rats. The contents of conjugated dienes and Schiff's bases increased by 68% and 70%, respectively, compared with control levels (Table 1). Activities of SOD and catalase decreased by 50% and 16%, respectively. However, the activity of G-6-PDH increased by 160% compared with the control.

The cold stress-induced activation of LPO increased the erythrocyte membrane permeability. This was confirmed by the increase in the concentrations of extraerythrocytic hemoglobin (by 63%) and iron (by 104%). Moreover, there was a 251% increase in the total plasma peroxidase activity compared with the control. This indicates destabilization of erythrocyte membranes in stress. Exogenous DSIP decreased the content of LPO products in erythrocyte membranes in the control. The concentrations of conjugated dienes and Schiff's bases in erythrocyte membranes decreased by 62% and 33%, respectively, compared with the control. DSIP decreased the content of LPO products in erythrocyte membranes compared with that in stressed rats. The concentration of conjugated dienes decreased by 35%. However, the concentration of Schiff's bases was 39% higher than the control level. Thus, cold stress activated LPO processes in erythrocyte membranes. DSIP decreased the intensity of LPO. This was probably due to a DSIP-induced increase in the activities of SOD and catalase in the erythrocytes of stressed animals to the control levels. DSIP increased the activity of catalase by 29% in the erythrocytes of stressed animals compared with the control.

In stressed DSIP-treated rats plasma concentrations of extraerythrocytic hemoglobin and iron were at the control levels, and total peroxidase activity decreased nearly to the control level. This indicates that DSIP stabilized the erythrocyte membranes in these animals. Obviously, in addition numerous effects mediated by the endocrine and nervous systems [1], DSIP can act directly on cell membranes. DSIP modifies physicochemical properties of membranes (the asymmetry and microviscosity and the composition and mobility of proteins and lipids), increases the membrane stability, changes its selective permeability, and inhibits the accumulation of LPO products.

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