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REACTIVITY OF MONONUCLEAR PHAGOCYTES IN THE LUNGS AND LIVER OF RATS EXPOSED TO LOW TEMPERATURES

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Resistance of the organism to stress, including cold stress, largely depends on reactivity of the mononuclear phagocyte system (MPS) [1, 4]. The MPS as a whole is a sufficiently heterogeneous cell population as regards both its morphological and functional characteristics and its role in various adaptive reactions [7]. In the investigation described below changes in some parameters of activity of MPS, and of its pulmonary and hepatic subdivisions, in the reaction of the organism to cold were studied.

EXPERIMENTAL METHOD

Experiments were carried out on 40 male Wistar rats weighing 160-180 g, divided into two groups: the animals of group 1 (n = 20) were kept at −7°C in a thermal chamber and rats of group 2 (n = 20) were kept at room temperature (20°C). The intensity of lipid peroxidation (LPO) in homogenates of the lungs and liver was determined after 2 and 24 h and after 8 days, by measuring accumulation of malonic dialdehyde (MDA) [9]. Protein was determined by Lowry's method. Alveolar macrophages were obtained by washing out the bronchopulmonary tract three times with heparinized Hanks' solution [8]; the total cell count was determined in a Goryaev's chamber. The washed out cell pool was deposited on a coverslip for differential counting, fixed with methanol, and stained with azure II-eosin. The ingestive power of the MPS was determined [5] by measuring the half-elimination time of colloidal carbon (from "Wagner") from the blood. The total number of macrophages loaded with carbon particles was counted in sections through the lungs and liver, stained with hematoxylin and eosin, under a magnification of 1000, and the total number of monocytes in the blood was counted.

EXPERIMENTAL RESULTS

After the animals had been kept for 2 h at a low temperature an almost twofold increase in the half-elimination time of colloidal carbon from the blood was observed (Table 1). Phagocytic activity of the Kupffer cells (KC) was reduced under these circumstances, whereas the phagocytic activity of the interstitial macrophages of the lungs was the same as in the control. The number of alveolar macrophages in the bronchopulmonary washings was reduced (Table 2). No LPO products were found to accumulate in the lung or liver tissues.

After exposure to cold during the next 24 h gradual disinhibition of MPS was observed, as reflected in a decrease in the half-elimination time of colloidal carbon from the blood, com-

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TABLE 1. Effect of Cooling on Activity of MPS and Content of LPO Products in Lungs and Liver (M ± m)

Experimental conditions	Half-elimination time of colloidal carbon, min	Phagocytic KC, %	Phagocytic macrophages in interstices of lungs, %	MDA concentration in homogenate, nanomoles/mg protein	
				lungs	liver
Control (n = 7)	4,2±0,1	76,7±1,5	24,0±1,4	1,1±0,07	1,2±0,09
Cooling					
2 h (*n*=7)	7,0±1,0*	52.8±1,4*	26,8±1,2	1,1±0,06	0,88±0,2
24h (*n*=7)	4,2±0,2	64,7±2,2	59,0±2,0*	2,1±0,09*	1,56±0,11
8days (*n*=7)	4,8±0,3	70,1±1,2	70,0±1,9**	2,0±0,2*	1,5±0,086*

Legend. Here and in Table 2: *P < 0.01, **P < 0.001, ***P < 0.05 compared with control. Number of animals given in parentheses.

TABLE 2. Changes in Number of Mononuclear Phagocytes in Lungs, Liver, and Peripheral Blood at Various Times during Cooling (M ± m)

Experimental conditions	Absolute number of monocytes in peripheral blood (× 10^3)	Number of KC per 1000 hepatocytes	Absolute number of monocytes in peripheral blood (× 10^3)
Control (n = 7)	6.4±0,5	338,4±13,5	0,5±0,04
Cooling			
2 h (*n*=7)	4,9±0,3***	339,4±22,3	0,28±0,03*
24 h (*n*=7)	8,0±0,6***	411,0±31,5*	1,06±0,1*
8days (*n*=7)	1,7±0,2**	387,0±19,0	0,99±0,095*

pared with that during exposure for 2 h. The number of KC undertaking phagocytosis of colloidal carbon was observed in the liver, although it did not reach the control level. Meanwhile in the lungs the number of interstitial macrophages, which had taken up carbon particles, was twice the control value. A tendency was noted for the total number of alveolar macrophages and of KC to increase. An increase in the number of monocytes was found in the peripheral blood, and an increase in the concentration of LPO products (MDA) in the lungs. This parameter in liver tissue by the end of the first day did not differ significantly from the control.

On the 8th day of exposure to cold the half-elimination time of colloidal carbon from the blood was at the control level (Table 1). The number of phagocytes with ingested carbon was increased, but the different levels of activity of the populations of these cells in the liver and lungs still remained. For instance, whereas the number of KC which phagocytosed carbon particles in the liver was restored only to the control level, in the lungs the number of interstitial macrophages with incorporated colloidal carbon was three times the initial number (Table 1). During this period the number of alveolar macrophages in the bronchopulmonary washings decreased and a high percentage of the cells thus obtained were alveolocytes (2.4 ± 0.2, compared with 1.0 ± 0.1 in the control). The MDA concentration in the lung tissues remained high. In the liver an increase in the concentration of this LPO product was found on the 8th day. The absolute number of monocytes was doubled.

Light-optical and electron-microscopic investigation of the lungs of animals kept at −7°C showed that during the first day acute disturbances of the hemodynamics and ventilation predominated. Later, especially toward the 8th day, definite activation of proliferative cellular reactions involving MPS and also the population of secretory alveolocytes was observed. Macrophages, located in the lumen of the alveoli, contained many heterogeneous inclusions, bounded by a membrane, formed as a result of fusion of primary lysosomes and phagosomes. Often membranous structures of surfactant and fragments of degenerating alveolar cells were seen in their cytoplasm.

The results of these experiments are evidence that the initial stages of exposure to cold are accompanied by a decrease in the ingestive power of cells of the MPS. This is associated with neuroendocrine adaptations arising in the acute phase of cold stress [3]. These changes are followed by activation of MPS, expressed more especially in the lung tissues. This different pattern of the response of the pulmonary and hepatic subdivisions of MPS may be due, first, to differences in metabolic specialization of the tissue macrophages and, second, to high reactivity of the lungs as a target organ for the action of low temperatures [2]. The difference is also reflected in the dynamics of accumulation of the products of LPO, and invariable component of stress-induced injury to membranous structures, in the lungs of the cooled animals. We know that free-radical destruction of cell membrane phospholipids is accompanied by the formation of highly reactive oxymetabolites: derivatives of L-hydroxy-5,8, 10,14-icosotetraenic acids. These compounds, even in extremely small quantities, can activate macrophages [6], and this could be one cause of activation of the pulmonary subdivision of MPS in our experiments. This same mechanism may lie at the basis of gradual disinhibition of KC. In fact, their ingestive power was not restored until the 8th day of exposure to low temperatures, when significant accumulation of MDA was detected.

The organ-specific orientation of the action of cold may thus be the cause of the formation of highly active metabolites of lipid peroxide nature in the lungs, capable even in the early stages of activating macrophages, and thereby bringing about heterogeneity in the manifestation of activity of the tissue phagocytes of the lungs and liver.

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