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ASSESSMENT OF THE STATE OF THE LUNG SURFACTANT SYSTEM IN HYPOXIA WITH PARAPULMONARY BLOOD OXYGENATION

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In the modern view, surface-active components of the phospholipid complex constituting the basis of the lung surfactant system (LSS) are responsible for the stability of the alveoli of the lungs. The study of LSS is important when the extrapulmonary gas exchange is maintained by means of membrane oxygenators (MO). There is reason to suppose that the "safety" of artificial support systems for the body can be judged to a certain extent from the degree of change in the LSS [3, 4].

The object of this investigation was to study the state of the LSS in animals with severe hypoxia of respiratory type, incompatible with life, and after arteriovenous connection to a Soviet "Sever-OMR" oxygenator for the purpose of treatment.

EXPERIMENTAL METHOD

Experiments were carried out on 18 mongrel dogs weighing 13-25 kg under morphine-hexobarbital anesthesia with muscle relaxation and artificial ventilation. The dogs were divided into three groups (with six animals in each group). The dogs of group 1 were provided with adequate artificial ventilation of the lungs. The animals of group 2 were on hypoventilation (respiration rate 3-4 breaths/min, respiratory minute volume 40% of normal); an arteriovenous shunt was created between the femoral artery and vein, with a volume velocity of blood flow of not more than 1 liter/min. The dogs of group 3 also were artificially hypoventilated. A type Sever-OMR MO was incorporated into the arteriovenous shunt [1]. The gas-exchange system functioned for 4 h.

The lung surfactant was investigated in broncho-alveolar washings and pieces of lung tissue. The washings were obtained by means of a modified double-barreled catheter of Fogarty

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type on the animals of group 1 by the end of the 3rd-4th hour of the experiment with normal artificial ventilation of the lungs, on group 2 in the terminal period immediately after the arterial blood pressure (BP) had fallen to 0 (after 40-90 min), and on the dogs of group 3 after operation of the MO for 4 h under hypoventilation conditions. Cold physiological saline was introduced through the catheter into the lungs and aspirated. The procedure was repeated three times. The total volume of washings was 60 ml. At the end of the experiment both lungs were removed from the thorax, weighed, and Pattle's coefficient (PC) of stability of the bubbles was determined.

The state of the LSS was evaluated by a combination of the following indices: 1) minimal and maximal surface tension (ST_{min} and ST_{max}) of the broncho-alveolar washings was determined by the plate detachment method on Wilhelmy's scales and the stability index (SI) was calculated by the formula $(ST_{max} - ST_{min})/(ST_{max} + ST_{min})$, 2) CP was determined in lung tissue and the lung coefficient (LC) was calculated by the equation: LC = (weight of lung)/(body weight)× 100; 3) the phospholipid composition of the broncho-alveolar washings was investigated by two dimensional thin-layer chromatography on silica-gel KSP after preliminary ultracentrifugation [3].*

The results were subjected to statistical analysis by the Mir-2 computer. Differences were considered to be significant at the $P < 0.05$ level.

EXPERIMENTAL RESULTS

The conditions of hypoventilation hypoxia chosen in these experiments led to the development of extremely severe metabolic and hemodynamic disturbances that were incompatible with life of the animals. Without treatment the dogs died 1-1.5 h after the beginning of the experiment. Connection to the MO enabled all the experimental animals to tolerate hypoxia for 4 h successfully.

Under conditions of acute respiratory insufficiency significant changes were observed in the surface-active properties of the broncho-alveolar washings compared with normally: ST_{min} was increased fivefold and the index of stability of the bubbles was reduced by half. Meanwhile ST_{max} and PC remained within normal limits (Table 1). After 4 h of parapulmonary oxygenation of the blood, against the background of the same hypoventilation no statistically significant changes were found in the surfactant properties of LSS compared with normal.

A study of the biochemical composition of the surface-active fraction of the broncho-alveolar washings from the lungs of the dogs of group 1 by two-dimensional thin-layer chromatography revealed seven phospholipid fractions under normal conditions, of which six fractions were identified: phosphatidylcholine (PC), phosphatidylethanolamine (PEA), lysophosphatidylcholine (LPC), phosphatidylinositol (PI), sphingomyelin (SPH), and phosphatidylserine (PS). The seventh (unidentified) fraction was evidently composed of polyglycerophosphatides. The relative percentages of the phospholipid fractions of LSS in the different groups of animals are given in Table 2.

In the animals of group 2 with parapulmonary blood oxygenation no significant differences from normal were found in any of the phospholipid fractions of the surfactant. The absence of changes in the level of the principal surface-active fraction of LSS (PC) during parapulmonary gas exchange with the Sever-OMR oxygenator suggests that the state of the LSS remained stable throughout the period of treatment.

The fall in the level of the PC fraction during hypoxia, accompanied by an increase in ST_{min} is evidence of profound disturbances of the function of the surfactant system. The cause of inactivation of the surfactant, it can be tentatively suggested, was an increase in activity of the enzyme phospholipase A_2 as a result of hypoxic injury to the cell membranes [2]. Indirect evidence in support of this view is given by the simultaneous increase in the level of fraction LPC (group 2) — the end product of conversion of PC through the action of phospholipase A_2. LPC is known to have weaker surface-active properties. There are also grounds for suggesting purely mechanical injury to the surfactant as a result of hypoxic disturbance of cell permeability and the appearance of interstitial and intra-alveolar edema.

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TABLE 1. Indices of Surface-Active Properties of Broncho-Alveolar Washings and of Lung Tissue during Normo- and Hypoventilation of the Lungs and Parapulmonary Blood Oxygenation

Group of animals	Statistical index	ST_{max}	ST_{min}	PC	LC	SI
		dynes/cm				
1	M	51,3	2	0,92	0,9	1,85
	$\pm m$	15,0	1	0,07	0,1	0,30
	P_{1-2}	>0,05	<0,05	>0,05	>0,05	<0,05
2	M	27,0	10	0,87	1,05	0,92
	$\pm m$	10,0	3	0,04	0,12	0,12
	P_{2-3}	>0,05	<0,05	>0,05	>0,05	<0,05
3	M	32,4	4	0,95	0,86	1,56
	$\pm m$	12,0	2	0,04	0,07	0,30
	P_{3-1}	>0,05	>0,05	>0,05	>0,05	>0,05

TABLE 2. Composition of Phospholipid Fractions (in %) of Bronchoalveolar Washings of Lung during Normo- and Hypoventilation of the Lungs and Parapulmonary Blood Oxygenation

Group of animals	Statistical index	PC	PEA	PI	LPC	SPH
1	M	78	16	0,80	4,03	0,70
	$\pm m$	1	3	0,04	0,30	0,04
	P_{1-2}	<0,05	>0,05	>0,05	<0,05	>0,05
2	M	68,0	12	1,0	10,5	0,5
	$\pm m$	1,9	2	0,8	2,2	0,1
	P_{2-3}	<0,05	>0,05	>0,05	<0,05	>0,05
3	M	79,9	14	0,83	3,03	0,8
	$\pm m$	0,9	3	0,17	0,50	0,2
	P_{3-1}	>0,05	>0,05	>0,05	>0,05	>0,05

The profound disturbances of the phospholipid composition of the surfactant and of its surface-active properties, characteristic of hypoxic states, did not develop when the MO was used therapeutically.

Inclusion of one of the first Soviet models of MO in the circuit of an arteriovenous shunt for the treatment of severe hypoxia for 4 h thus caused no disturbances of LSS and created conditions under which no hypoxic changes developed in the lungs or in the rest of the body. This distinguishes the MO as superior to other types of gas-exchange systems, the use of which is attended by serious disturbances of the LSS [4, 5].

These experiments revealed definite correlation between the PC concentration and surface-active properties of the LSS. On this basis the writers recommend the use of the relatively simple and rapid method of determination of ST_{min} of the lung washings in order to monitor adequacy of the physiological state of the LSS during prolonged parapulmonary blood oxygenation. The results obtained by this method can form the basis of a "safety test" when the new method of treatment of acute respiratory insufficiency, by parapulmonary blood oxygenation with membrane oxygenators, is introduced into clinical practice.

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