EFFECT OF ALCOHOL POISONING DURING PREGNANCY ON THE LUNG SURFACTANT SYSTEM OF NEWBORN RATS

A. K. Zagorul'ko, E. E. Fisik, and E. N. Tikus

UDC 616.24-008.4-092.9:612.212.014.1.014.462.8]-02:618.3-06:[615.917:547.262]-055.26

KEY WORDS: lung surfactant; alcohol

There is much experimental evidence to show that under the influence of alcohol, acting on the mother during pregnancy, the newborn animals develop a symptom-complex known as the fetal alcohol syndrome (FAS) [3]. The FAS is characterized by a triad of typical signs: delayed physical and mental development, and malformation of the cranial and facial bones [2]. Meanwhile no information could be found in the accessible literature on the effect of alcohol, acting on the mother during pregnancy, on the state of the lung surfactant system (LS) of fetuses and newborn animals, although we know that during the first few days after birth, children whose mothers suffer from alcoholism develop respiratory failure [2]. Since changes in the surfactant properties of LS play an important role in the pathogenesis of various pathological states of the lungs, the aim of this investigation was to study the state of the LS system of newborn rats whose mothers had alcohol poisoning.

EXPERIMENTAL METHOD

Material for study consisted of the lungs of 45 newborn rats whose mothers were eight Wistar albino rats weighing from 290 to 350 g, which were given a 50% solution of ethyl alcohol, in a dose of 4 ml/kg body weight, on alternate days including the whole period of pregnancy, for 4-4.5 months. As a rule the young were born at night. All the young rats were born alive. The animals were killed on the 1st day of life by decapitation. The control series consisted of 16 newborn rats whose two mothers were healthy.

The surface-active properties of SL were studied by physicochemical (measurement of surface tension — ST — of the surface-active fraction of the lung extracts isolated by differential centrifugation) and biochemical (determination of total lipids, phospholipids of SL, followed by thin-layer chromatography of the latter) methods.

To study the cellular component of the SL system (type II alveolocytes, alveolar macrophages) electron microscopy was used. For this purpose pieces of lungs were fixed in 2.5% glutaraldehyde solution in phosphate buffer and then dehydrated in alcohols of increasing concentration and absolute acetone, and embedded in epoxide resins. Ultrathin sections were cut on an LKB ultramicrotome (Sweden), stained with lead citrate, and examined in the UEMV-100K electron microscope.

EXPERIMENTAL RESULTS

The lungs of animals of the experimental group weighed less than the lungs of the young control rats: 189.6 ± 7.3 and 242.8 ± 14.3 mg respectively ($p < 0.01$).

The results obtained by a study of the surface-active properties of SL of the newborn rats are given in Table 1.

It follows from Table 1 that in newborn rats whose mothers before and during pregnancy were exposed to the action of ethyl alcohol, the surface-active properties of SL were inhibited. This is shown by the significantly higher level of ST_{min} of the surface-active fraction of lung extracts than in the control and the lower value of Clements' index of stability (IS). In animals of the experimental group the concentrations of total lipids and phospholipids of SL were significantly reduced. Inhibition of the surface activity of SL was evidently connected with reduction of the content of the phosphatidylcholine fraction, with the highest

Department of Pathological Anatomy, Crimean Medical Institute, Simferopol'. (Presented by Academician of the Academy of Medical Sciences of the USSR A. P. Avtsyn.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 110, No. 8, pp. 142-144, August, 1990. Original article submitted June 1, 1989.

TABLE 1. Values of ST_{min}, Clements' IS, and Total Lipids, Phospholipids of SL and Their Qualitative Composition of the Surface-Active Fraction of Lung Extracts from Newborn Rats Whose Mothers Had Alcohol Poisoning

Group of rats	ST_{min}', mN/m	IS	Concentration of total lipids, g/liter	Concentration of phospholipids		
				total, mmoles/liter	made up of	
					Phosphatidylcholine, %	Phosphatidylethanolamine, %
Experimental (n = 45)	19,1±2,8	0,85±0,05	0,95±0,006	0,028±0,004	58,0±2,4	17,3±1,2
Control (n = 16)	11,6±1,2	1,30±0,02	1,09±0,001	0,048±0,004	67,7±2,0	8,8±1,2
p	<0,02	<0,001	<0,001	<0,05	<0,01	<0,01

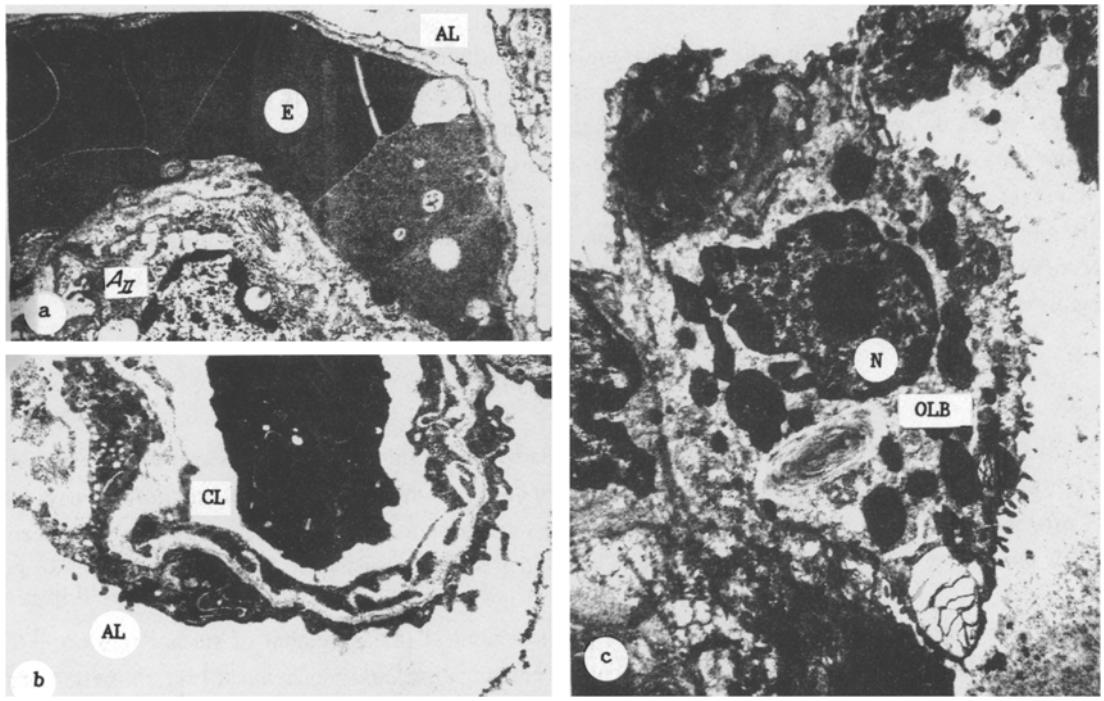

Fig. 1. Ultrastructural changes in components of ABB of newborn rat lungs following alcohol poisoning of their mothers. a) Congestion of capillaries of alveolar septa. E) Erythrocytes in capillary lumen, AL) alveolar lumen, AII) type II alveolocyte? 16,000×; b) widening of interstitial space (IN), AL) alveolar lumen, CL) capillary lumen. 13,000×; c) Increase in number of OLB in type II alveolocyte. N) Nucleus. 18,000×.

surface activity, in the composition of phospholipids of SL ($p < 0.01$). This state of affairs, and also the fact that the fraction of phosphatidylethanolamine, a precursor of phosphatidylcholine, and methylation of which is the main pathway of phosphatidylcholine formation in the fetal lungs, was appreciably increased in the composition of the phospholipids of SL, may indicate that inhibition of the surface-active properties of SL is connected with a disturbance of synthesis of the surface-active substance in the type II alveolocytes. This conclusion is confirmed also by the results of the electron-microscopic investigations.

At the same time the possibility of a direct damaging effect of ethanol on SL cannot be ruled out, because ethanol is a lipotropic substance and can pass freely through the placental barrier from mother to fetus.

The results of the electron-microscopic investigation indicate that changes are present in the lungs of newborn rats, including those affecting all components of the air—blood barrier (ABB). A characteristic feature of the lungs of newborn animals is the marked congestion of the capillaries of the alveolar septa (Fig. 1a). In some areas, stasis of erythrocytes was observed in the capillaries. Endothelial cells as a rule preserved their characteristic structure with a central nucleus-containing part, and with a paranuclear arrangement of organelles, and with long cytoplasmic processes, spreading over the well contoured basement membrane. In the peripheral zones of the endothelial cells there were numerous micropinocytotic vesicles. The inter-

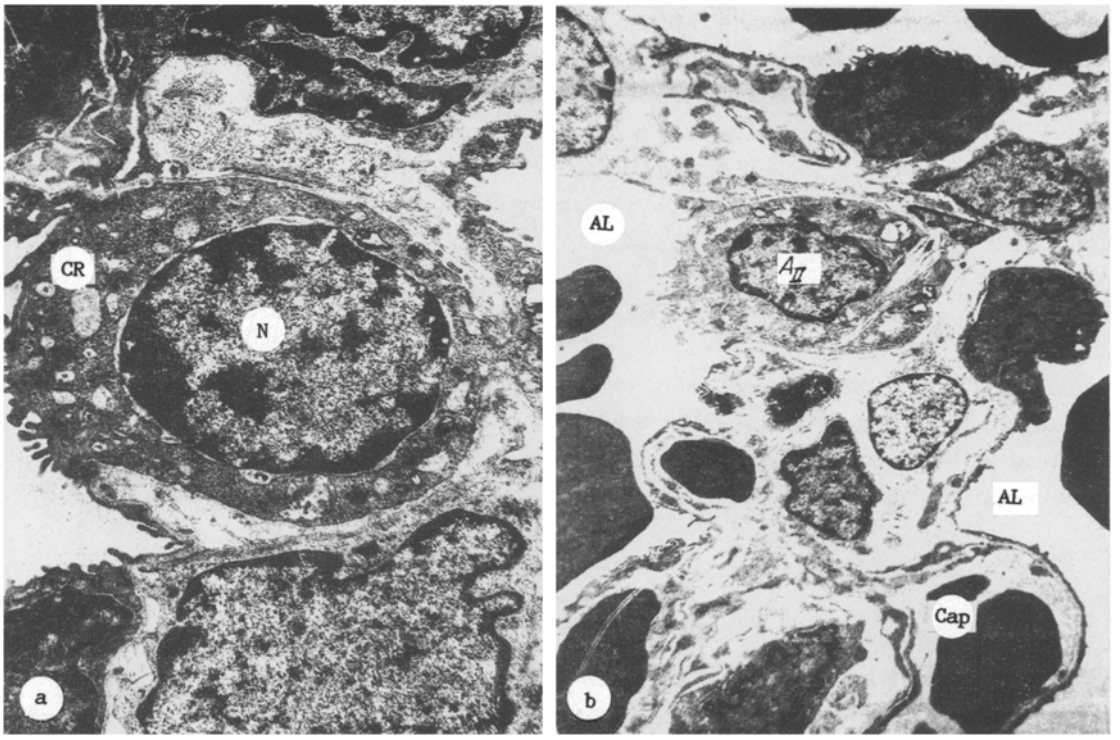

Fig. 2. Ultrastructural changes in components of ABB of lungs of newborn rats whose mothers had alcohol poisoning. a) Dilatation of tubules of cytoplasmic reticulum (CR) in a "pale" type II alveolocyte. N) Nucleus. 13,000×; b) Erythrocytes in alveolar lumen (AL). CAP) Capillaries in alveolar septa. AII) Type II alveolocyte. 7000×.

stitial space was widened over a considerable extent of the ABB because of edema (Fig. 1b). Changes in the opposite direction were found in cells of the alveolar epithelium,

Most of the hypertrophied type II alveolocytes had a round nucleus, sometimes with festooned edges. The cytoplasm of the cells had high electron density, distinguishing them as "dark" type II alveolocytes. They contained a complex of developed organelles, among which mitochondria with densely packed cristae and osmiophilic lamellar bodies (OLB), numbering 12 to 14 per cell, could be distinguished (Fig. 1c). Moderately well defined microvilli could be seen on the apical surface of the cells.

At the same time, besides these "dark" type II alveolocytes, "pale" cells could also be seen in the young rats' lungs. These "pale" type II alveolocytes were characterized by a round nucleus with their chromatin more or less uniformly distributed throughout the karyoplasm, and a paler cytoplasm of low electron density, in which a few small mitochondria and OLB, present in relatively small numbers (four or five per cell), were dominant. A characteristic feature of these cells was the presence of dilated tubules of the cytoplasmic reticulum (Fig. 2a).

The presence of "dark" and "pale" cells in the alveolar epithelium has been described [1] in the proliferative phase of development of regeneration and compensatory hypertrophy of the lung. The appearance of hypertrophied type II alveolocytes is associated in this case with compensatory growth of the lung after partial resection of the organ.

In the experiment now being described, the increase in functional activity of the type II alveolocytes with the appearance of hypertrophied forms of these cells may be connected with the need to produce extra SL in order to make good its deficiency, arising on the one hand, as was stated above, from the direct damaging action of ethanol on the surface-active film, and on the other hand, from the inactivating action of the blood plasma proteins on SL, which penetrate into the alveolar lumen because of increased permeability of the components of ABB. This last hypothesis is indirectly confirmed by the fact that during electron-microscopic investigation, besides intensification of microvesicle formation in the peripheral zones of the endothelial cells of the capillaries of the alveolar septa and widening of the interstitial space on account of its edema, serous fluid and large numbers of blood cells, mainly erythrocytes, are found in the alveolar lumen (Fig. 2b).

Thus the results are evidence that inhibition of the surface-active properties of SL takes place in newborn rats whose mothers were poisoned with alcohol before and during pregnancy, reduction of the surface activity of SL is evidently linked both with the direct damaging action of ethyl alcohol, which passes freely through the placental barrier, on the surface-active film and also with inactivation of the surface-active substance by blood plasma proteins, entering the alveolus on account of increased permeability of the components of ABB. Inhibition of the surface-active properties of SL under these circumstances is accompanied by enhanced functional activity of the type II alveolocytes, responsible for SL production, with the appearance of hypertrophied forms of these cells.

LITERATURE CITED

1. L. K. Romanova, Cellular Bases of Regeneration in Mammals [in Russian], Moscow (1984), pp. 40-62.
2. Ya. P. Sokol'skii, M. L. Tarakhovskii, P. G. Leshchinskii, and L. A. Barkov, Effect of Alcohol on Mother, Fetus, and Child [in Russian], Kiev (1988).
3. L. V. Timoshenko, N. P. Skakun, and G. K. Skakun, The Fetal Alcohol Syndrome [in Russian], Kiev (1987).

EFFECT OF TEMPERATURE ON STATE OF THE SMALL INTESTINAL MICROCIRCULATION DURING ACUTE ISCHEMIA

V. S. Shinkarenko, J. Collantes-Jara, and O. S. Kulikova

UDC 616.341-005.4-07:616.16-031:611.341]-008.1-092:612.59

KEY WORDS: microcirculation; ischemia; hyperthermia; hypothermia

Preservation of the viability of an intestinal loop after total or partial interruption of its blood flow (thrombosis, compression of the vessels) is largely dependent on the state of the microcirculation, and under clinical conditions, besides other methods, this may be improved by the use of Kerte's method, namely by heating the ischemic portion of the intestine. Meanwhile there is evidence that a high temperature has an unfavorable effect on the state of the intestine [1] and, conversely, that its blood supply is improved by lowering the temperature [1-3]. However, the character of features in the microcirculation has not been studied under these conditions.

The aim of this investigation was to study pathophysiological mechanisms determining the state of the microcirculation in acute local ischemia of the small intestine under normo-, hyper-, and hypothermic conditions.

EXPERIMENTAL METHOD

The investigation was conducted on 72 male Wistar rats weighing from 170 to 320 g, under pentobarbital anesthesia (6 mg/100 g body weight, intramuscularly). Ischemia of a portion of the small intestine was produced by applying a ligature for 1 h to the base of a loop of intestine, exteriorized through an incision in the abdominal wall, on the light guide of a microscope. In control experiments, only eventration of the intestinal loop was carried out, without application of a ligature.

The investigation was carried out during reperfusion of the ischemic part of the intestine for 60 min after resumption of the blood flow under normo-, hyper-, and hypothermic conditions (at 38, 42, and 20°C respectively). The necessary temperature

Laboratory of General Pathology of the Microcirculation, and Laboratory of Experimental Pathomorphology, Research Institute of General Pathology and Pathological Physiology, Academy of Medical Sciences of the USSR, Moscow.

(Presented by Academician of the Academy of Medical Sciences of the USSR G. N. Kryzhanovskii.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 110, No. 8, pp. 145-148, August, 1990. Original article submitted July 7, 1989.

0007-4888/90/0008-1034$12.50