CLARIFICATION FACTOR AND THE COURSE OF DEVELOPMENT OF EXPERIMENTAL SILICOSIS IN RATS

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Numerous investigations have shown that in normal conditions during alimentary hyperlipemia the lungs play an active part in the clarification of the lipemic serum, by retaining and splitting the lipids entering them [4-6, 12]. The role of the lungs in clarifying the lipemic serum is seen to particular advantage after removal of part of the organ [9].

In atherosclerosis, characterized by chronic hyperlipemia, a fall in the activity of the clarification factor is observed [7, 8]. Inderbitzen [11] associates this fall with a disturbance of the lipopexic function of the lung, with the exhaustion of its lipopexic reserves in conjunction with the manifestations of chronic hyperlipemia. This state was produced by Inderbitzen in experiments on dogs receiving a prolonged high-fat diet. The fatty infiltration of the lung tissue in such animals was confirmed histochemically.

In the author's previous experiments [1-3] a considerable accumulation of lipids was found in the lungs of rats after injection of various dusts into this organ. The increase in the content of lipids in the experimental animals (by comparison with the controls) was clearly apparent within a few days after introduction of the dust into the lungs, and it continued along with the development of the silicotic process, as histochemical investigations confirmed.

On the basis of previous findings [1] indicating an increase in the lipopexic and a decrease in the lipolytic activity of the lungs of rats with silicosis, it might be expected that the activity of the clarification factor is decreased in experimental silicosis.

The object of the present investigation was to study the state of the clarification factor in silicosis.

EXPERIMENTAL METHOD

Experiments were carried out on 20 albino rats (male) weighing 180-200 g, kept in the ordinary vivarium conditions.

In healthy animals, 48 h after the first determination of the clarification factor, administration of quartz dust took place by intratracheal injection (50 mg in 1 ml physiological saline), and further determinations of the clarification factor were made 5, 14, 15, 60, 180, and 270 days layer. At the end of the experiment 15 rats were still alive, 1 died between the 1st and 6th, and 4 between the 6th and 9th months after injection of the dust. Blood for analysis was taken from the tail 30 min after intraperitoneal injection of heparin in a dose of 50 units/100 g body weight. Blood from each animal in a volume of 0.1 ml was transferred into the incubation medium, consisting of an emulsion of 2% peach oil in 5% human serum albumin solution in Sörensen's phosphate buffer, pH 8.0. The emulsion was prepared by mechanical shaking in an emulsifier for 40 min. The size of the oil droplets in the emulsion did not exceed about 1 μ. Samples were kept for 2 h in a thermostatically controlled water bath at 37±0.5°, in which they were shaken at the rate of 120 oscillations/min. Control samples of emulsion without blood were incubated in the water bath at the same time as the experimental samples.

The activity of the clarification factor was determined from the amount of higher nonesterified fatty acids (HNFA) formed after incubation for 2 h, and expressed in meq/ml on the basis of the difference between the HNFA content of 1 ml of the test sample (emulsion + blood) and of the emulsion alone. The method was suggested by the Department of Pathophysiology (Head, Professor S. M. Leites) of the Central Postgraduate Medical Institute.

The degree of development of the silicotic process at the time of the last investigation (270 days after administration of dust) in 10 rats was assessed from the weight of the dry lungs, the content of quartz dust in the lungs

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Clarification Factor in the Blood of Rats before and after Intratracheal Injection of Quartz Dust (M± σ)

Index	Before injection of dust	Time after injection of dust (in days)					
		5	14	15	60	180	270
Clarification factor	7,1±2,2 (20)	5,9±1,2 (20)	3,5±1,2 (20)	2,5±0,58 (20)	6,5±1,6 (19)	5,3±0,95 (19)	4,0±1,1 (15)
P		<0,05	≪0,001	≪0,001	>0,1	<0,01	≪0,001

Note. Number of animals in parentheses.

was determined by the method of Stacey and King [13], the hydroxyproline content by Chvapil's method [10], and the lipids by the method described earlier [2]; histochemical investigations were also carried out on lung sections stained with hematoxylin-eosin and by Van Gieson's method.

EXPERIMENTAL RESULTS AND DISCUSSION

As the results of the histological investigation showed, by the end of the 9th month of the experimental period the development of silicotic changes in the animal's lungs had attained a considerable degree. In all the investigated rats numerous typical silicotic nodules were found, in various phases of maturation, consisting in some cases of almost cell-free structures with a dense network of coarse collagen fibers. Many nodules formed extensive confluent conglomerates. In some places perivascular and peribronchial sclerosis were well defined. The lungs were emphysematous.

The mean content of hydroxyproline in the lungs was 8.96 mg (compared with the normal value of 4-5 mg for rats of the same age) and the content of lipids was 131.5 mg (normally 25-35 mg). The mean content of quartz dust found in the lungs was 24.6 mg.

As the table shows, a significant decrease in the activity of the clarification factor (by comparison with the initial level) was found in the course of development of silicosis at all stages of the experiment except the 60th day.

The fall in the activity of the clarification factor observed in most of the animals 5 days after administration of dust continued until the 14th or 15th day, at which time it was observed in all the rats. By the 60th day, all the rats showed a tendency for restoration of the normal activity of the clarification factor, and its activity in a series of animals reached its initial level. At the later stages of development of silicosis (180 and 270 days after injection of dust) the clarification factor again showed a diminished activity by comparison with the initial level. The most marked fall in its activity was found on the 14th and 15th days after administration of dust, and thereafter the activity was at a higher level.

The author [1] previously found a similar pattern of changes in the lipolytic activity of the lung and liver tissues of rats sacrificed in groups 5, 14, 60, 180, and 270 days after the injection of quartz dust. The similarity of the character of the changes in the activity of the clarification factor and the lipolytic activity in experimental silicosis probably points to the identity of the enzyme entering the blood stream from the lungs after injection of heparin. The lowered content of this enzyme in the tissues in silicosis leads to a fall in the activity of clarification factor in this pathological state.

During the investigation of the lipopexic function of the lungs in earlier experiments, the author suggested that with the development of silicosis the reserves of the "fat depot" of the lungs were exhausted, so that with alimentary fat loading the content of lipids in the lungs of rats receiving injections of dust increases more than in healthy animals only in the earliest stages of the experiment. Later the difference disappears, and 6 or 9 months after administration of dust, fat loading in general ceases to have a visible effect on the now high content of lipids in the lung tissue. Taking into consideration the data of Inderbitzen, cited above, it may be concluded that the "fat loading" of the lungs caused by silicosis is a supplementary mechanism leading to a lowering of the activity of the clarification factor.

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