Low Serum Alpha-1-Antitrypsin Level as a Contributory Factor of Combined Emphysema in Silicosis

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The First Department of Internal Medicine, Tohoku University School of Medicine, Sendai, Rosai Hospital for Silicosis, Fujiwara-cho, Shiota-gun, Tochigi Pref., and The First Department of Internal Medicine,† Yamagata University School of Medicine, Yamagata*

TOKITA, K., IKEDA, K., KANNO, K., SAITO, K., CHIYOTANI, K. and NAKAMURA, T. *Low Serum Alpha-1-Antitrypsin Level as a Contributory Factor of Combined Emphysema in Silicosis.* Tohoku J. exp. Med., 1975, 117 (1), 61-67 — In order to evaluate low serum alpha-1-antitrypsin level as a contributory factor of combined emphysema in silicotic patients, serum alpha-1-antitrypsin analysis was carried out in 80 patients with silicosis. Low serum alpha-1-antitrypsin level was found in 5 patients. Large opacities were observed roentgenologically in only 1 out of these 5 cases in contrast to 31 of the other 75 cases. Also the suggestive findings for the emphysema were showed in 4 of these 5 cases while such findings were found on their chest X-ray films in only 43 of the other 75 cases. FEV_{1.0}% below 50 were calculated in 3 of the 5 cases, on the contrary in 22 of the other 75 cases. RV above 50% predicted value was showed by all the 5 low antitrypsin patients, in contrast to only 25 of other 74 cases. The silicotic patients with low serum alpha-1-antitrypsin concentration are most likely to have an association with a high incidence of complicated emphysema. ——— alpha-1-antitrypsin; emphysema; silicosis; trypsin inhibitory capacity; immunodiffusion

Silicosis is a occupational disease, caused by inhalation of dusts containing free silicic acid, characterized by proliferation of connective tissues in the lung. It exhibits several pathological patterns. The chest X-ray figure of silicosis is characterized by disseminated nodular shadows, although additional large opacities may be found in some of the cases. Some of the silicotic patients have emphysematous changes in their lung, but others do not show such changes.

Our interest has been directed toward the factors which may play a role to cause complicated emphysema in certain cases of silicosis. Stimulated by recent theories (Eriksson 1965; Talamo 1971) that the intrapulmonary proteolytic enzyme might play a role in the pathogenesis of chronic obstructive lung disease, serum alpha-1-antitrypsin level was determined in 80 cases of silicosis, and the serum alpha-1-antitrypsin concentration was correlated with roentgenological

signs suggestive of emphysema and with the results of pulmonary function tests. Our observation suggested that the low serum alpha-1-antitrypsin level was associated with a high incidence of emphysema complicated with silicosis.

SUBJECTS AND METHODS

Subjects were 80 patients with silicosis excluding silicotuberculosis, hospitalized in the Rosai Hospital for Silicosis. They were all male patients, aged from 41 to 78 years old (average 61.2 years), having occupational careers as metal miner, stonemason, tunnel constructor, or as industrial workers in foundry, in glass factory, in pottery and so on.

The average ages when the patients were first diagnosed as silicosis necessary to be hospitalized were 52.5, ranged from 29 to 72 years. The chest X-ray patterns of the patients were classified according to the international criteria. It is difficult to discriminate emphysema combined with silicosis on the chest X-ray films, and we noted the lowered diaphragm (below 11th thoracic vertebra in the back) and bullous shadow as a sign suggesting of emphysema. Vital capacity (VC) and one second forced expiratory volume ($FEV_{1.0}$) were determined by means of a spirometer, and $FEV_{1.0}\%$ was calculated as $FEV_{1.0}/VC \times 100$. Pulmonary diffusing capacity was determined at rest by the carbon monoxide steady state method (Bates et al. 1955), and % DLCO was expressed as a percentage of the values predicted on the basis of age and stature. Residual lung volume (RV) was determined by the helium dilution method (Meneely and Kaltreider 1949) and compared to the predicted value for normal persons.

Blood samples were withdrawn from the patients, and the serum was stored below -25°C until use. Serum trypsin inhibitor capacity was determined by Eriksson's method (1965). Serum alpha-1-antitrypsin level was determined by a single radial immunodiffusion method on commercially purchased agar plate (M-partigen plate, Behringer A.G.). Acid starch gel electrophoresis was performed by the method after Fagerhol and Braend (1965).

RESULTS

Fig. 1 is a scattergram of the values of trypsin inhibitor capacity versus immunologically determined serum alpha-1-antitrypsin level in 80 cases of silicosis. There was a statistically significant correlation between these two values ($r=0.54$, $p<0.01$).

Our previous work showed that the mean $\pm$ s.d. of serum trypsin inhibitor capacity in normal subjects was 1.01 ± 0.08 mg trypsin/ml, and the level at 0.70 mg trypsin/ml as considered as a critical level of alpha-1-antitrypsin. With respect to the trypsin inhibitor capacities of 80 patients with silicosis examined, those of 73 were within normal limits. And those of other 7 patients showed significantly lower concentrations (0.57 to 0.70 mg trypsin/ml), which might be termed intermediate level. But the patients who seemed to have the homozygous deficient geno-type were not found in our group.

By a single radial immunodiffusion method, serum alpha-1-antitrypsin level in normal subjects was found to be 290 ± 60 (s.d.) mg/100 ml, and the levels lower than 200 mg/100 ml were considered to be significantly low values. In this study 6 patients with significantly low values were found out of 80 patients examined, with the range from 150 to 180 mg/100 ml.

By an acid starch gel electrophoresis, usual type, MM, was found in 78 out of 80 cases. MS type was in only 2 cases, and these 2 belonged to the low serum

Fig. 1. Relationship between serum trypsin inhibitory capacity and serum alpha-1-antitrypsin concentration determined by the single radial immunodiffusion method on 80 cases of silicosis. Regression line is expressed as $Y=277.3X+31.6$. Both values show highly significant correlation ($r=0.54$, $p<0.01$).

alpha-1-antitrypsin group by both Eriksson's method and the radial immunodiffusion method.

We defined 5 of 80 cases of silicosis as group A, showing low alpha-1-antitrypsin level by both Eriksson's method and the radial immunodiffusion method, so that the group A included not only genetically determined phenotype, showing intermediate level, but also usual MM type with low serum alpha-1-antitrypsin concentration in some reason. Roentgenological and functional comparison was made on the group A and group B which covered other 75 cases.

Although the average ages of the group A and the group B were essentially the same (62 years versus 61 years), average ages when they were required for the treatment in hospital were considerably different (46 years versus 53 years). From these results, it might be considered that the pathological conditions of the patients in the group A seemed to progress more rapidly than those of the group B, because average ages when they were employed in the metal mine, etc., were almost the same.

Table 1 showed the chest X-ray patterns of the groups A and B classified according to the international criteria for silicosis. In the group A only small nodules (p, m, n) were shown in 4 of 5 cases (80%), and large opacities (A, B, C) were shown in only 1 case (20%). On the contrary, in the group B small opacities were observed in 44 cases (59%), and large in 31 cases (41%). Table 2 shows roentgenological findings concerning the association of emphysema. The

TABLE 1. *Differences of chest X-ray pattern between Groups A and B of the silicosis*

Chest X-ray pattern	Group A		Group B	
	Number of cases	Total	Number of cases	Total
1 p	0	0	2	2
m	0		0	
n	0		0	
2 p	0	4	9	23
m	1		10	
n	0		4	
3 p	1	3	7	19
m	1		4	
n	1		8	
4 A	0	1	19	31
B	1		9	
C	0		3	

Abbreviations: 1 (p, m, n), 2 (p, m, n), 3 (p, m, n), 4 (A, B, C); international criteria for chest X-ray pattern of silicosis.

TABLE 2. *Incidences of silicosis with or without emphysema in chest X-ray*

	Group A	Group B
	Number of cases (%)	Number of cases (%)
With findings of emphysema	4 (80.0)	43 (57.3)
Without finding of emphysema	1 (20.0)	32 (42.6)
Total	5 (100.0)	75 (100.0)

emphysematous changes were observed in 4 of 5 cases (80%) of the group A, whereas in 43 of 75 cases (57%) of the group B. These results lead us to consider the association of the group A with emphysematous changes and of the group B with large opacities.

The results of the pulmonary function tests were tabulated in Table 3. The severe air way obstruction was considered to be present in these cases whose FEV_{1.0} % was less than 50%, and it was found in 3 of 5 cases (60%) of the group A, while in 22 of 75 cases (29%) in the group B. Vital capacity, which was expressed as a percentage of the predicted value on the basis of sex, age, and stature, showed no difference between the two groups. All of the 5 cases examined in the group A fell in residual volume of 51 to 70%, but 49 of 74 cases in the group B showed residual volume of $\leq 50\%$. So that the patients of the group A seemed to have a higher residual volume than those of the group B. As to diffusing capacity no significant difference was observed between these two groups. These pulmonary function tests indicated that patients of the group A had lower FEV_{1.0}% value and

TABLE 3. *Values of pulmonary function of Group A and Group B*

Percentage to the predicted standard value (VC, RV, DLCO) or percentage to VC (FEV _{1.0})	Number of cases							
	VC		RV		FEV _{1.0}		DLCO	
	A	B	A	B	A	B	A	B
$\leq 50.0\%$	0	10	0	49	3	22	0	12
50.1-70.0%	3	19	5	24	1	29	3	27
70.1-90.0%	2	26	0	1	0	23	0	21
$\geq 90.1\%$	0	20	0	0	1	1	2	12
Total	5	75	5	74	5	75	5	72

Abbreviations: VC, vital capacity; RV, residual volume; FEV_{1.0}%, ratio of one second forced expiratory volume/vital capacity; DLCO, diffusing capacity for carbon monoxide.

higher residual volume than the group B. It might be concluded, therefore, that the patients of the group A were more prone to associate with emphysema than those of the group B.

DISCUSSION

Although silicosis is a kind of pulmonary fibrosis caused by inhalation of dusts containing free silicic acid, individual patients' conditions may also modify their pulmonary changes. In some cases of silicosis, small granulomas confluent each other to build a large conglomerate lesion in the lung. It might be conceivable that individual conditions, for instance, the proliferative tendency of connective tissue either locally or generally, might contribute to it.

On the other hand, some of the silicotic patients show emphysematous changes in their lung to a certain extent. Because the emphysema accelerates to deteriorate pulmonary function of the silicotic patients, we are looking for the factors contributing to it. Since Laurell and Eriksson (1963) first reported the relationship between inherited serum alpha-1-antitrypsin deficiency and pulmonary emphysema, numerous lines of evidence have documented that not only the deficiency but also intermediate level (Fagerhol and Hauge 1969) of this serum protein could associate the pulmonary emphysema significantly. To understand this relationship, the following hypothesis may be presented that various kinds of proteolytic enzymes released from the leukocyte, macrophage and bacteria into the pulmonary tissue can digest the structural protein in the lung, if antiprotease activity is reduced, for instance, in the case of lowered alpha-1-antitrypsin level in the serum. Really the alpha-1-antitrypsin was shown to inhibit not only trypsin (Schwick et al. 1966) but also chymotrypsin (Schwick et al. 1966), bacterial protease (Talamo 1971), neutral proteases and elastase from lysosomal granules of neutrophils (Knepper and Bean 1966; Janoff 1970), skin collagenase (Pierce et al. 1969), plasmin, and thrombin (Gans and Tan 1967). Recently two binding sites were found to be present on alpha-1-antitrypsin (Cohen 1973). Ward and Talamo (1973) presented that the serum deficient in alpha-1-antitrypsin is also deficient in

chemotactic factor inactivator. Deficiency of this anti-factor also takes share in the pulmonary changes by imbalance in the generation and control of chemotactic factors, leading inordinate delivery of neutrophils and their enzymes in inflammatory exudate.

The purpose of this paper is to elucidate whether this relationship is applicable to the emphysema combined to silicosis. Several methods have been proposed for the determination of the serum alpha-1-antitrypsin and it is advised that one should employ two or more methods based on the different principles. In this work we estimated serum alpha-1-antitrypsin level with Eriksson's method and the single radial immunodiffusion method. There was a fairly good statistic correlation between values determined with these two methods. But there were some exceptional cases which showed the low value with one method but the value within normal limit with the other method. Excluding uncertain cases, we picked up 5 cases showing low serum alpha-1-antitrypsin levels with both methods.

One difficulty in this work is how to diagnose the silicosis with emphysema. We employed the roentgenological and functional criteria. Of course they do not coincide exactly with each other. Recently Felson (1973) described that the minimal or moderate pulmonary emphysema was not possible to diagnose roentgenologically, but the advanced pulmonary emphysema was detected by a decreased excursion of the diaphragm, diminished vascularity of the central and peripheral portion of the lung, and an increased retrosternal clear space. But in the case of silicosis, it is more difficult to delineate the combined emphysema, because emphysema is not always generalized but may also be localized and silicotic opacities darken the pulmonary area. Taking note of the bullous shadow and the subordinate position of the diaphragm as a suggestive sign of emphysema, we could confirm the association of low alpha-1-antitrypsin group with roentgenological emphysematous changes.

Of pulmonary function tests, values of $FEV_{1.0}\%$ and $\%RV$ are thought to be significant, reflecting changes for emphysema. These values in our silicotic patients also suggested the association of low serum alpha-1-antitrypsin with pulmonary emphysema.

Our results suggested that the silicotic patients with low serum alpha-1-antitrypsin were necessary to be observed carefully by the doctors. Workers with a low serum alpha-1-antitrypsin level, once exposed to the dusty environment, were seemingly to show a rapid trend to develop emphysema combined with silicosis, their lung function becoming worse followed by severe downwards clinical course. It is considered to be reasonable that man with a low serum alpha-1-antitrypsin level has to be advised not to work in the dusty environment and the serum alpha-1-antitrypsin level should be examined before employment.

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