

BIOLOGICAL EFFECTS OF MINERAL DUSTS. IN VITRO AND IN VIVO STUDIES

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ABSTRACT

Illite and, among kaolins, kaolin Sz with particles smaller than $2\ \mu\text{m}$, cause a tissue reaction of the "storage" type in the rat lung that is similar to the reaction due to bentonite found in former investigations⁸. Kaolin Sz with particles smaller than $5\ \mu\text{m}$ causes a mixed, partly "storage", partly "foreign body" reaction. The surface activity of the dusts which produce a "storage" reaction - in our investigations determined by methylene blue adsorption - is of high intensity. The TTC-RA₅₀ activity of these dusts is also distinctly marked.

Kaolin S with its mineral features deviating from kaolin Sz and its low surface activity causes a "foreign body" reaction.

In relation to the "storage" reaction caused by aluminium silicates, the adsorption, very probably the albumin adsorption, is of fundamental importance. In our investigations we evaluated the harmful effects of several aluminium silicates *in vivo* and *in vitro* by means of the methylene blue adsorption and the TTC-RA₅₀ value, taking into account the mineral features and composition of the rocks and dusts.

In animal experiments with coal dusts from local silicosis-endangered coal mines⁹ we noticed as early as 1962, that the effect of dust cannot be evaluated only by means of its quartz content and that the presence or absence of free quartz surfaces is essential for a specific collagenous effect. In 1965 in animal experiments with several bentonite dusts⁸ we found that bentonite when it contains higher amounts of quartz, induces "storage" type reaction in the rat lung.

We demonstrated^{5,7} that various aluminium silicates like montmorillonite or kaolin, when they occur together with quartz or are experimentally added to it, inhibit the specific effect of quartz. Our investigations proved that aluminium silicates added to quartz take part in the development of a tissue reaction. Silicosis, enameller's pneumoconiosis and bentonitosis occur with a different frequency in mines with the same quartz content.

In several publications^{2,6,7} we reported that we could notice in the experiments (haemolysis, TTC-RA etc.), carried out *in vitro*, the toxicity of different dusts without finding any parallel between the examinations *in vitro* and *in vivo*. The dusts occurring in mineral mines are different in their mineral

composition and in their particle size: we examined the effects of bentonite, illite and several kaolin dusts as well as of coal and corundum dusts on the rat lung *in vivo* and on the rat peritoneal macrophages *in vitro*. In addition to that, we determined the adsorption of methylene blue.

The purpose of our investigations was to find out to what extent the mineral composition and/or the particle size affect the biological effects *in vivo* and *in vitro*, and also, to what extent the *in vivo* effect of dust can be determined by these investigations.

METHODS AND MATERIALS

Dust samples were administered to rats intratracheally in a quantity of 40 mg. The rats were killed 5, 15, 40 and 365 days after administration, the lungs were removed and inspected. The distribution of particles in dust samples was determined by means of the particle size counter (TGZ).

TTC-RA₅₀ values were determined by the method of Marks and James³; the methylene blue adsorption was measured by the method of Juhász¹. The data about the examined dusts are summarized in Table 1.

TABLE 1
Data of the examined dust samples.

Designation	Composition	Particle size (μm)
Quartz H with HCl treated	Hochenbuch silica sand	< 5
Kaolin S	18% quartz + 82% kaolin	< 3
Kaolin Sz	5% quartz + 95% kaolin	< 2; 2-5
Illite F	Illite	< 2; 2-5
Bentonite I	3% quartz + 18% amorph cristobalite + 3% feldspar + 76% montmorillonite	< 2; < 5
Corundum	90% aluminium oxide + 10% quartz	< 5
Coal M	95% coal + 5% quartz	< 5 (92%)
Coal T	Coal dust	< 5 (92%)

RESULTS AND DISCUSSION

The tissue reactions observed in the rat lung were the following: bentonite and illite with particles smaller than 5 μm (regardless of the particle size) caused tissue reactions of the "storage" type; kaolin Sz with particles smaller than 2 μm caused reactions of the "storage" type, but with the particles between 2 and 5 μm it induced a mixed reaction in which the "storage" type prevailed over the "foreign body" type; kaolin S caused tissue reactions of the "foreign body" type.

The TTC-RA₅₀ values, the methylene blue adsorption and the types of tissue reaction are demonstrated in Table 2.

TABLE 2
TTC-RA₅₀ value, methylene blue adsorption, and type of tissue reaction in the rat lung.

Sample	TTC-RA ₅₀ μg/7 × 10 ⁶ cells	Methylene blue adsorption (mg/g)	Type of tissue reaction
Quartz H - < 5 μm	100 ± 11	3 ± 1	Progressive fibrosis
Kaolin S - < 3 μm	200 ± 19	17 ± 1	"Foreign body"
Kaolin Sz - < 2 μm	120 ± 10	30 ± 6	"Storage"
Kaolin Sz - 2-5 μm	200 ± 14	24 ± 4	Mainly "storage", rarely "foreign body"
Illite F - < 2 μm	40 ± 6	103 ± 10	"Storage"
Illite F - 2-5 μm	100 ± 9	54 ± 7	—
Bentonite I - < 2 μm	30 ± 5	300 ± 19	"Storage"
Bentonite I - < 5 μm	60 ± 5	269 ± 15	"Storage"
Corundum - < 5 μm	> 1000	2 ± 1	"Foreign body"
Coal M - < 5 μm (92%)	750 ± 22	10 ± 3	"Foreign body"
Coal T - < 5 μm (92%)	800 ± 30	20 ± 4	"Foreign body"

The *in vivo* "storage" reaction produced by aluminium silicates (illite, kaolin Sz with particles smaller than 2 μm, bentonite) with a methylene blue adsorption capacity of 30 mg/g or much higher caused a TTC-RA₅₀ of 120 μg or lower. However, kaolin which adsorbed 17 mg/g of methylene blue and caused a TTC-RA₅₀ of 200 μg led to a "foreign body" reaction. The methylene blue adsorption of corundum which produces a "foreign body" reaction as well as the coal dusts of the M- and T-type was less than 20 mg/g and the TTC-RA₅₀ values were above 700 mg. The mineral composition can help in evaluation. The four examined aluminium silicates differ in their mineral composition. On the basis of their different mineral characteristics and substitutional ability the dusts can be ranged according to their expected "surface activity", from the highest to the lowest: bentonite - illite - kaolin Sz - kaolin S⁴. In our experiments the methylene blue adsorption was one feature of the surface activity which decreased in the above mentioned sequence (Table 2). TTC-RA₅₀ decreased, too. The particle size influences the methylene blue adsorption and the TTC-RA₅₀ reaction to a small extent that can be neglected compared with the differences in the mineral characteristics. We suppose that in the case of aluminium silicates the range of adsorption capacity - first of all the albumin adsorption is important - takes probably a leading part in the development of the "storage" reaction. The methylene blue adsorption capacity of other dusts was generally of low range. The toxic effect to be expected is characterized by the TTC-RA₅₀ reaction. In the case of quartz, coal and corundum dusts a connection between the TTC-RA₅₀ value and the *in vivo* effect can be observed. The low range TTC-RA₅₀ value signals that a foreign body reaction has to be expected. Our investigations

support again, and even more emphatically, the statement that the biological effect of dusts is influenced by their mineral features and composition.

To evaluate the biological effect it is essential to identify the origin of the rocks, to know the mineral features of rocks and dusts (morphology, surface, composition etc.), their chemical composition and the relations between quartz and its accompanying minerals and substances.

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