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FROM KELLER AND HECKMAN 2024344654

P. 2

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**CABOT**

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April 13, 1999

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Re: Notice of TSCA Section 8(e) Substantial Risk Information for [ ]

Dear Sir or Madam:

Cabot is submitting this letter as notice under the Toxic Substances Control Act (TSCA) section 8(e) regarding substantial risk information. We have determined that the results of an acute inhalation study conducted in 1994 on Cabot dimethyldichlorosilane, reaction products with silica, [ ] (CAS No. 68611-44-9) are reportable based on the stringent guidelines for reporting acute inhalation studies contained in the U.S. EPA June 1991 Section 8(e) Reporting Guide. The reported LC<sub>50</sub> value falls into the extremely toxic range (i.e., LC50 < 0.5 mg/L). On April 2, 1999, a self-disclosure notice was filed with U.S. EPA Region I on Cabot's behalf by John Dubeck of Keller and Heckman, LLP; in follow-up, this section 8(e) letter is being submitted within 15 working days of realizing that the study might represent a section 8(e) reportable discovery.

[TSCA Confidential Business Information deleted.]

[ ] was tested in a 4-hour acute inhalation toxicity study in [ ] in accordance with EPA Guidelines for Test Procedures, Subdivision F, Series 81-3 and TSCA 40 CFR 796.1150. Five male and 5 female rats were exposed to aerosol atmospheres of [ ] at 0.21, 0.54, or 2.1 mg/L. All animals in the 2.1 mg/L exposure group and 5/5 males and 2/5 females in the 0.54 mg/L exposure group died during the exposure period. No lethality was observed in the lowest exposure group. The calculated LC<sub>50</sub> and 95% confidence limits were reported to be 0.45 mg/L (0.33 - 0.62). [ ]

Whether or not these results represented a new discovery in 1994 is unclear. Findings of lethality at comparable concentrations for similar products have been reported in the literature (see table below). Lethality was observed in a number of repeated exposure inhalation studies in which rats were exposed to lower concentrations (i.e., < 0.45 mg/L) of compounds similar to [Cabot's product]. Reuzel et al. (1991) reported lethality in rats exposed to [Compound A] at 0.209 mg/L during a 14-day range finding study. IUCLID (1996) reported other studies in which lethality was observed. In one study, "spontaneous" lethality was reported in 60/235 rats exposed for 4 hours/day for up to one year to [Compound B] at 0.08 mg/L. In another 1-year study, lethality was reported in rats exposed for 5 hours/day to [Compound C] at 0.05 mg/L. [ ]

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APR-13-1999 2:59PM

FROM MILLER AND HECKMAN 2024344654

P. 3

APR-13-1999 23:00

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**CABOT**

TSCA 8(e) Coordinator

April 13, 1999

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Product	Type of inhalation study	Lowest concentration at which lethality was observed (mg/L)	Reference
[Cabot's product]	4-hour	0.54 (LC <sub>50</sub> = 0.45)	Cabot (1994)
[Compound A]	14-day	0.209 <sup>a</sup>	IUCLID; Reuzel (1991)
[Compound B]	1-year	0.08 <sup>a</sup>	IUCLID
[Compound C]	1-year	0.05 <sup>a</sup>	IUCLID

<sup>a</sup> Note: the length of exposure that resulted in lethality is not known.

These studies identify lethality at air concentrations similar or lower to those observed in our study. However, there are some limitations to interpreting these studies since the details on exposure time and resulting lethality have not been provided. In other words, it is unclear if lethality occurred following exposure to only one 4-hour exposure or to repeated short-term exposures. There is some evidence that lethality in the other studies occurred early on, as in the [Compound C] study. More specifically, the exposure to [Compound C] was reportedly reduced to 2 days/week as a result of unexpected lethality. Although these data suggest lethality following an acute exposure, they do not definitively make clear whether the findings are attributable to single or multiple exposures.

In summary, Cabot's data are, in principle, consistent with the results of studies previously presented in the public domain. However, the lack of detail presented in the publicly-available study summaries leaves open the possibility that our results represent the lowest concentration to produce lethality after a single 4-hour exposure.

If you have any questions, please do not hesitate to contact me at 978-670-6965.

Sincerely,

*D. Cooper Rees*

D. Cooper Rees, Ph.D., DABT  
 Director of Toxicology and Corporate Toxicologist

Enclosures [TSCA Confidential Business Information - not included]

cc: Rosina Toscano, U.S. EPA - Region I  
 John Dubeck, Esq., Keller and Heckman, LLP

**References**

IUCLID. 1996. International Uniform Chemical Information Database, Edition 1, Existing Chemicals - 1996. European Commission Joint Research Centre. European Chemicals Bureau.

Reuzel, P. G. J., Bruijntjes, J. P., Feron, V. J., and Weutersen, R. A. 1991. Subchronic inhalation toxicity of amorphous silicas and quartz dust in rats. *Ed Chem. Toxic.* 29(5):341-354.

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## SUBCHRONIC INHALATION TOXICITY OF AMORPHOUS SILICAS AND QUARTZ DUST IN RATS

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(Accepted 21 February 1991)

**Abstract**—The inhalation toxicity of three amorphous silicas (Aerosil 200, Aerosil R 974 and Sipernat 22S) was compared with that of quartz dust. Rats were exposed to 1, 6 or 30 mg Aerosil 200/m<sup>3</sup>, 30 mg Aerosil R 974/m<sup>3</sup>, 30 mg Sipernat 22S/m<sup>3</sup> or 60 mg quartz/m<sup>3</sup> for 6 hr/day, 5 days/week for 13 wk. Some rats were killed at the end of the exposure period and some were killed 13, 26, 35 or 52 wk after the end of exposure. Clinical signs, body weight, haematology, biochemistry, histology, organ weights, retention of test material in the lungs and regional lymph nodes, collagen content of the lungs, and gross and microscopic pathology were determined in order to disclose possible adverse effects and to study the reversibility, stability or progression of the effects. All test materials induced increases in lung weight, and pulmonary lesions such as accumulation of alveolar macrophages, inflammation, alveolar bronchiolization and fibrosis. In addition, rats exposed to Aerosil 200, Aerosil R 974 or quartz developed granulomatous lesions. Silicosis was observed only in quartz-exposed animals. At the end of the exposure period, Aerosil 200 and quartz had induced the most severe changes. Quartz dust was hardly cleared from the lungs and the changes in the lungs progressed during the post-treatment period, and eventually resulted in lesions resembling silicotic nodules and in one squamous cell carcinoma. Although Aerosil 200 was very quickly cleared from the lungs and regional lymph nodes, the changes in these organs were only partly reversed during the post-exposure period in rats exposed to 30 mg/m<sup>3</sup>. Aerosil R 974 and the lower levels of Aerosil 200 resulted in less severe, and mostly reversible, changes. The slightest changes were found after exposure to Sipernat 22S, notwithstanding the persistence of this silica in the lungs during the major part of the post-treatment period. The results of this study revealed that only quartz induced progressive lesions in the lungs resembling silicotic nodules. Of the amorphous silicas examined Aerosil 200 induced the most severe changes in the lungs, which only partly recovered, whereas Sipernat 22S induced the least severe, completely reversible lung changes.

### INTRODUCTION

Amorphous silicas are widely used in synthetic resins, plastics, lacquers, vinyl coatings, varnishes, pharmaceuticals, cosmetics, adhesives, paints and foods and thus there is widespread occupational exposure to these substances (Groth *et al.*, 1979). Exposure levels during production were monitored between 1974 and 1986 and varied from 13 mg total dust/m<sup>3</sup> in 1982 to 2.2 mg/m<sup>3</sup> in 1984 and 4.4 mg/m<sup>3</sup> in 1986 (Degussa AG, 1983 and 1987).

Amorphous silicas have the same basic chemical formula (almost 100% SiO<sub>2</sub>) as quartz, a crystalline silica. It is well known that quartz dust can induce silicosis, a chronic lung disease characterized by severe fibrosis and granulomas in the lungs (Gleason *et al.*, 1969; Reiser *et al.*, 1982; Reiser and Last, 1979 and 1983; Renne *et al.*, 1985). Amorphous silicas, however, have been reported in animal studies to have no (Gärtner, 1952) or little (Groth *et al.*, 1979; Klosterkötter, 1965; Schepers *et al.*, 1957) fibrogenicity. No silicosis was found in workers exposed to amorphous silica for up to 34.4 years and no definite relationships between exposure to Aerosil (an amorphous silica) and symptomatic complaints were found (Degussa AG, 1987).

To verify the difference in effects on the lungs between quartz and three amorphous silicas

(Aerosil 200, Aerosil R 974 and Sipernat 22S) we carried out a 13-wk inhalation study followed by post-exposure observation periods of up to 1 yr.

Aerosil 200, a hydrophilic silica, was selected as the primary test material because it is the most widely used grade and it has a propensity to generate airborne particles. Aerosil R 974 was selected because it is produced by a chemical treatment of Aerosil 200: a carbon content of approximately 1% transforms this material into the hydrophobic state. Sipernat 22S was chosen because it has the same specific surface area as Aerosil 200. The concentrations selected were based on the results of 2-wk concentration-finding studies with each of the test materials. The results of these 2-wk studies have been summarized in the present paper.

Aerosil 200 was examined at three exposure levels. From the 2-wk studies we knew that 17 mg Aerosil 200/m<sup>3</sup> induced very slight adverse effects in the lung. Therefore, the highest exposure level (30 mg/m<sup>3</sup>) could be expected to induce clear effects but no mortality. The level of 6 mg/m<sup>3</sup> was selected because it was the German TLV value for inert dust. Both Aerosil R 974 and Sipernat 22S were examined at one level only (30 mg/m<sup>3</sup>) which was chosen so that we could compare the effects of the three amorphous silica dusts. Quartz had induced only slight changes after 2 wk of exposure at a level of 70 mg/m<sup>3</sup>. In order to be sure that changes would occur, quartz dust was examined at a level of 60 mg/m<sup>3</sup>.

\*To whom correspondence should be addressed.

## MATERIALS AND METHODS

*Concentration-finding studies*

2-Wk inhalation toxicity studies were carried out with Aerosil 200 (0, 17, 44 or 164 mg/m<sup>3</sup> air), Aerosil R 974 (0, 31, 87 or 209 mg/m<sup>3</sup> air), Sipernat 22S (0, 46, 180 or 668 mg/m<sup>3</sup> air) and quartz dust (0, 70, 211 or 901 mg/m<sup>3</sup> air).

SPF-bred Wistar rats (Cpb: WU, 4 wk old) were purchased from the TNO Central Institute for the Breeding of Laboratory Animals, Zeist, The Netherlands. Groups of 10 males and 10 females were housed in exposure chambers (see below) and exposed to atmospheres containing one of the test compounds for 6 hr/day, 5 days/wk for 2 wk. Body weights, food consumption, haematological parameters, organ weights and gross and microscopic pathology were used to examine the toxic potential of the test materials.

*Sub-chronic study*

*Animals and maintenance.* 6-Wk-old male ( $n = 490$ ) and female ( $n = 490$ ) SPF-bred Wistar rats (Cpb: WU, Wistar random) were purchased from the TNO Central Institute for the Breeding of Laboratory Animals. They were housed singly in stainless-steel wire cages in Hazleton H 1000 inhalation chambers throughout the whole 13-wk exposure period. The chambers were kept at 21–23°C, and 65–75% relative humidity, with an air-flow of approximately 40 m<sup>3</sup>/hr. The rats were provided *ad lib.* with unfluoridated tap-water and fed the Institute's stock diet for rats. During exposure the rats were deprived of food and water. After the exposure period those rats that were retained for the post-treatment periods were transferred from the inhalation chambers to an animal room and housed in wire-mesh, stainless-steel cages, five males and five females to a cage.

*Test material.* Commercial grade amorphous silicas were manufactured by Degussa AG (Frankfurt am Main, Germany) and quartz dust (Sikron F 300) was obtained from Quartz Werke (Frochen, Germany). The specifications and typical values of the tested silicas are presented in Table 1.

The very small primary particles (<6-about 45 nm, calculated as the arithmetic mean of transmission electron micrograph magnifications) form agglomerates and aggregates (Plate 1). Because of the weakness of the bonds and the electrostatic charge of the particles it was impossible to determine the

Table 2. Mean concentrations of test materials in test atmospheres

Test material	Target concn (mg/m <sup>3</sup> )	Actual concn (mg/m <sup>3</sup> ± SEM)
None (controls)	—	—
Aerosil 200	17	1.3 ± 0.1
	44	5.9 ± 0.2
	30	31.0 ± 0.9
Aerosil R 974	30	34.7 ± 0.7
Sipernat 22S	30	34.9 ± 0.5
Quartz	60	58.5 ± 0.7

aerodynamic agglomerate/aggregate size distribution in the test atmospheres.

The range of the geometric agglomerate/aggregate size distribution was 1  $\mu$ m about 120  $\mu$ m for the amorphous silicas with maxima at about 10 and 100  $\mu$ m. Quartz particle sizes globally varied between 0.1 and 25  $\mu$ m (Table 1).

Aerosols were generated using the Institute's dust generators, which were composed of a dust feed mechanism and an atomizer operated by compressed air. The concentrations of test material in the test atmospheres were determined by gravimetry. Samples of the test atmospheres were drawn through glass fibre filters (Sartorius SM 13430). The filters were weighed just before and after sampling.

*Study design and treatment.* There were six test groups and one control group each containing 70 males and 70 females. The rats were exposed for 6 hr/day, 5 days/wk for 13 wk to concentrations of test material as presented in Table 2. After the exposure period and 13, 26, 39 and 52 wk after exposure 20, 10, 10, 10 and 20 rats/sex/group, respectively, were killed.

*Observations, haematology and biochemistry.* Clinical observations were made daily. Body weights were recorded weekly during the exposure period and once every 4 wk thereafter. Haematological and urinary parameters were determined in 10 rats/sex/group at 13-wk intervals. Haematological determinations include cell counts, haemoglobin content, packed cell volume, white-cell counts, differential white-cell counts, prothrombin time, thrombocytes, albumin, alkaline phosphatase, alanine aminotransferase, aspartate aminotransferase, urea, total protein, creatinine, total bilirubin, calcium, potassium, sodium, inorganic phosphate, cholesterol and glucose. Urinary parameters included appearance, volume, density and pH, analysis for protein, occult blood, glucose and ketones and microscopy of sediment.

Table 1. Specifications of the amorphous silicas and quartz

Characteristic	Aerosil 200	Aerosil R 974	Sipernat 22S	Quartz
Structure	Amorphous	Amorphous	Amorphous	Crystalline
BET-surface area (m <sup>2</sup> /g)	200	170	150	<1.5
Behaviour against water	Hydrophilic	Hydrophobic	Hydrophilic	Hydrophilic
Mean primary particle size ( $\mu$ m)	12	12	18	8500
Primary particle shape	Spherical	Spherical	Spherical	Coarse, irregular sharp edges
Agglomeration size ( $\mu$ m)	NA	NA	7	No agglomerations
pH value	3.6–4.3	3.6–4.3	6.3	7
SiO <sub>2</sub> content (%)	>99.8	>99.8	98	99
Sodium sulphate (%)	NA	NA	0.3	NA
Chloride (ppm)	<250	<300	NA	NA

NA = not applicable

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Inhalation toxicity of amorphous silica

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**Pathology.** From 50% of the rats killed at each stage the following tissues and organs were collected and preserved in 4% aqueous, neutral phosphate buffered formaldehyde solutions: lungs, mediastinal and hilar lymph nodes, trachea, larynx, adrenals, aorta, axillary lymph nodes, brain (brainstem, cerebrum and cerebellum), caecum, coagulating glands, colon, duodenum, epididymides, eyes, heart, ileum, jejunum, kidneys, liver, mammary glands, mesenteric lymph nodes, nose (nasal cavity), oesophagus, ovaries, pancreas, parathyroids, parotid salivary glands, pharynx, pituitary, prostate, rectum, seminal vesicles, skeletal muscle (thigh), skin/subcutis (flank), spinal cord, spleen, sternum with bone marrow, stomach, sublingual salivary glands, testes, thymus (if identifiable), thyroid, urinary bladder and uterus. The lungs, adrenals, brain, heart, kidneys, liver, spleen, testes and thymus were weighed. The lungs were fixed by intratracheal inflation with the fixative under 10-cm water pressure. Tissues required for microscopic examination were embedded in Paraplast, sectioned at 5  $\mu$ m and stained with haematoxylin and eosin. Histopathological examination was carried out on all tissues and organs collected at the end of the exposure period, and on the respiratory tract and regional lymph nodes collected after 13, 26, 39 and 52 wk of observation.

**Collagen content in lungs and silicon content in lungs and associated lymph nodes.** From the other 50% of the rats killed at each of the stages one lung was assigned for determination of hydroxyproline as an indicator of the collagen content in the lungs. The lung tissue was hydrolysed in 6N-HCl at 140°C for 16 hr. After filtration of the solution, the hydroxyproline content was determined by means of a Technicon Autoanalyzer II, using the Industrial Method NL 142-78F (Technicon Instruments, 1978). A conversion factor of 8 was used to estimate the collagen content from the amount of hydroxyproline. The total collagen content in both lungs was calculated from the collagen content determined in the one lung, the weight of that lung and the weight of both lungs, assuming a similar collagen content in both lungs. The total collagen content was expressed relative to body weight in order to correct for differences in lung weight.

To study the clearance of the silicas from the lungs the amount of silicon was determined in the remaining lung and in the associated lymph nodes at the end of the exposure period and at 3-month intervals thereafter. The amounts of silicon found in one lung were converted on the basis of the separate lung weights to values for both lungs, assuming that the distribution of the silica particles between the two lungs was proportional to the relative weights of the two lungs. Silicon content was determined by flame absorption spectrometry (Lichte *et al.*, 1980).

**Statistical analyses.** Body weights were analysed by an analysis of co-variance (Cochran, 1957) followed by the Dunnett's multiple comparison test (Dunnett, 1955). Analysis of variance (Steel and Torrie, 1960) followed by the Dunnett's multiple comparison test were applied to the organ weights, and haematological and biochemical data. Incidences of histopathological changes were analysed by the Fisher exact probability test (Siegel 1956).

## RESULTS

*Concentration-finding studies*

**Aerosil 200.** Respiratory distress was observed in all groups. One female exposed to 164 mg/m<sup>3</sup> died during the study. Body-weight gain and mean food consumption were reduced in males exposed to 44 or 164 mg/m<sup>3</sup> in comparison with the controls. Histopathology revealed increased septal cellularity, alveolar interstitial pneumonia and granulomas in the lungs of males and females of all test groups. There was a clear concentration-response relationship.

It was concluded that the no-observed-adverse effect level of Aerosil 200 was lower than 17 mg/m<sup>3</sup>.

**Aerosil R 974.** Respiratory distress was observed in all rats exposed to Aerosil R 974. Four males and two females exposed to 209 mg/m<sup>3</sup> air died. Body-weight gain and food consumption were lower in males and females exposed to 87 or 209 mg Aerosil R 974/m<sup>3</sup> than in controls. Haemoglobin content, packed cell volume and red blood cell count were elevated in male rats exposed to 87 or 209 mg Aerosil R 974/m<sup>3</sup>. Females in the highest dose group exhibited elevated packed cell volumes.

Absolute and relative lung weights were dose-relatedly increased in all test groups. Moreover, the lungs of several animals in all test groups were pale, spotted, swollen and spongy. Microscopic examination revealed a concentration-related increase in cellularity, alveolar oedema and granulomas of the lungs of all test groups. It was concluded that the no-observed-adverse effect level of Aerosil R 974 was lower than 31 mg/m<sup>3</sup>.

**Sipernat 225.** Respiratory distress was observed in all groups exposed to Sipernat. One male exposed to 680 mg/m<sup>3</sup> died. In comparison with the controls, body-weight gain was reduced in males exposed to 170 or 680 mg/m<sup>3</sup> and in females exposed to 680 mg/m<sup>3</sup> and mean food consumption was decreased in rats exposed to 170 or 680 mg/m<sup>3</sup>. Haemoglobin content, packed cell volume and red blood cell counts were higher in males exposed to 170 or 680 mg/m<sup>3</sup> than in controls.

Absolute and relative lung weights were dose-relatedly increased in all test groups. The lungs of several rats exposed to the highest dose were spotted and swollen and had an irregular surface. Microscopic examination revealed increased septal cellularity, interstitial pneumonia and accumulations of particulate material in the lungs of most males exposed to 170 or 680 mg/m<sup>3</sup> and in females of all test groups. Granulomas were found in the lungs of rats exposed to 680 mg/m<sup>3</sup>. It was concluded that the no-observed-adverse effect level of Sipernat 225 was lower than 46 mg/m<sup>3</sup> air.

**Quartz dust.** Body-weight gain was reduced in males exposed to 211 or 911 mg/m<sup>3</sup>. Mortality did not occur and food consumption and haematology were unremarkable. Absolute and relative lung weights were increased in rats exposed to 901 mg/m<sup>3</sup>. Histopathology revealed increased cellularity, interstitial pneumonia and accumulations of alveolar macrophages in lungs of most rats exposed to quartz dust.

Table 3. F weights of rats exposed to amorphous silica or quartz for 13 wk and then observed for up to 52 wk

Weeks after exposure†	Body weights (g) of rats exposed to						
	Control	Aerosil 200			Aerosil R 974 (30 mg/m <sup>3</sup> )	Sipernat 225 (30 mg/m <sup>3</sup> )	Quartz (60 mg/m <sup>3</sup> )
		1 mg/m <sup>3</sup>	6 mg/m <sup>3</sup>	30 mg/m <sup>3</sup>			
<b>Males</b>							
-13	109 ± 2 (70)	107 ± 1 (70)	106 ± 1 (70)	106 ± 1 (70)	108 ± 2 (70)	108 ± 1 (70)	106 ± 1 (70)
0	380 ± 5 (70)	378 ± 6 (70)	369 ± 4 (70)	355 ± 5* (70)	380 ± 4 (70)	363 ± 5* (70)	376 ± 4 (70)
13	441 ± 7 (50)	443 ± 6 (50)	437 ± 6 (50)	423 ± 6 (50)	446 ± 6 (45)	428 ± 6 (49)	412 ± 6* (50)
26	492 ± 8 (40)	495 ± 8 (40)	487 ± 8 (40)	480 ± 9 (40)	504 ± 8 (35)	484 ± 8 (39)	457 ± 7** (40)
39	516 ± 14 (20)	524 ± 11 (20)	511 ± 14 (20)	509 ± 14 (20)	516 ± 12 (20)	494 ± 12 (19)	467 ± 10 (20)
52	530 ± 17 (20)	534 ± 14 (20)	528 ± 15 (20)	535 ± 14 (20)	536 ± 12 (20)	509 ± 13 (18)	465 ± 12* (20)
<b>Females</b>							
-13	107 ± 1 (70)	106 ± 1 (70)	109 ± 1 (70)	107 ± 1 (70)	105 ± 1 (70)	107 ± 1 (70)	105 ± 1 (70)
0	227 ± 2 (70)	231 ± 2 (70)	233 ± 2 (70)	226 ± 2 (70)	229 ± 2 (70)	223 ± 3 (69)	226 ± 2 (70)
13	254 ± 3 (50)	258 ± 3 (50)	261 ± 3 (50)	259 ± 3 (50)	254 ± 2 (50)	253 ± 3 (49)	246 ± 3 (50)
26	274 ± 5 (30)	285 ± 5 (30)	283 ± 6 (30)	288 ± 6 (29)	280 ± 4 (29)	284 ± 6 (25)	258 ± 5 (30)
39	295 ± 8 (20)	298 ± 7 (20)	306 ± 9 (20)	317 ± 10 (19)	286 ± 7 (18)	311 ± 10 (19)	278 ± 7 (20)
52	315 ± 8 (19)	312 ± 7 (20)	318 ± 9 (20)	335 ± 11 (19)	301 ± 9 (17)	334 ± 14 (18)	278 ± 9* (18)

†Wk -13 = start of the exposure period.

Values are means ± SEM for the numbers of rats indicated in parentheses.

Values marked with asterisks differ significantly (covariance and Dunnett's tests, two-sided) from the corresponding control value (\*P < 0.05; \*\*P < 0.01).

**Subchronic study.**

**Exposure concentrations.** The actual exposure concentrations of the test material (Table 2) were quite close to the target concentrations in spite of difficulties presented by the electrical charge on the particles, which was not allowed to be neutralized.

**Clinical signs, body weights, haematology and clinical chemistry.** During exposure, there was a concentration-related increase in the respiration rate of animals exposed to Aerosil 200. The respiration rate quickly returned to normal when the exposure was ended.

At the end of the exposure period, body-weight gain was 5-10% lower in males exposed to 30 mg Aerosil 200/m<sup>3</sup> or 30 mg Sipernat 225/m<sup>3</sup>. The body weight of quartz-exposed rats was not affected during the exposure period. After a 13-wk post-exposure period body weights had returned to normal in all groups exposed to the amorphous silicas. However, the quartz-exposed rats showed a slightly progressive reduction in weight gain throughout the post-exposure period (Table 3).

At the end of the exposure period, neutrophilic leucocyte counts were higher in most groups than

in controls, but the elevation was only statistically significant in rats exposed to 30 mg Aerosil 200/m<sup>3</sup> or to quartz. These counts were back to normal in rats exposed to Aerosil 200 within 13 wk of termination of the exposure. In quartz-exposed rats the neutrophilic leucocyte counts increased during the first 13 wk after the end of the exposures and remained high during the whole post-exposure period (Table 4).

Red blood cell counts, haemoglobin content and packed cell volumes had slightly increased in males exposed to 30 mg Aerosil 200/m<sup>3</sup>, Aerosil R 974 or quartz by the end of the exposure period. After 13 wk of non-exposure these parameters had returned to normal in all animals exposed to amorphous silica. The males exposed to quartz continued to show high red blood cell values throughout the observation period.

From 13 wk after exposure, alanine aminotransferase activities increased in quartz-exposed rats. In males the increase was approximately 50-90% (P < 0.01) in comparison with the controls. Alkaline phosphatase activity had increased in rats exposed to quartz 52 wk after the exposure period. The remaining haematological and biochemical parameters that were examined did not show differences

Table 4. Neutrophilic leucocyte count in rats exposed to amorphous silica or quartz for 13 wk and then observed for up to 52 wk

Group	Neutrophilic leucocyte count (10 <sup>9</sup> /litre) in post-exposure wk:				
	0	13	26	39	52
<b>Males</b>					
Controls	20 ± 3	14 ± 4	20 ± 3	15 ± 2	31 ± 6
Aerosil 200 (30 mg/m <sup>3</sup> )	44 ± 6**	21 ± 2	17 ± 2	20 ± 3	22 ± 2
Aerosil R 974 (30 mg/m <sup>3</sup> )	26 ± 3	19 ± 3	20 ± 2	14 ± 3	20 ± 3
Sipernat 225 (30 mg/m <sup>3</sup> )	28 ± 2	21 ± 4	24 ± 9	24 ± 3	19 ± 3
Quartz (60 mg/m <sup>3</sup> )	48 ± 4**	94 ± 12**	73 ± 10**	75 ± 4**	95 ± 14**
<b>Females</b>					
Controls	12 ± 1	13 ± 2	12 ± 3	13 ± 2	14 ± 2
Aerosil 200 (30 mg/m <sup>3</sup> )	35 ± 6**	21 ± 2	19 ± 2	9 ± 1	19 ± 3
Aerosil R 974 (30 mg/m <sup>3</sup> )	24 ± 1	31 ± 2	12 ± 2	17 ± 5	17 ± 3
Sipernat 225 (30 mg/m <sup>3</sup> )	25 ± 4	18 ± 3	12 ± 2	15 ± 2	22 ± 4
Quartz (60 mg/m <sup>3</sup> )	31 ± 7*	47 ± 4**	55 ± 8**	54 ± 5**	58 ± 11**

Values are means ± SEM for groups of 10 rats, and those marked with asterisks differ significantly (ANOVA and Dunnett's test, two-sided) from the corresponding control value (\*P < 0.05; \*\*P < 0.01).

Libalation toxicity of amorphous silicas

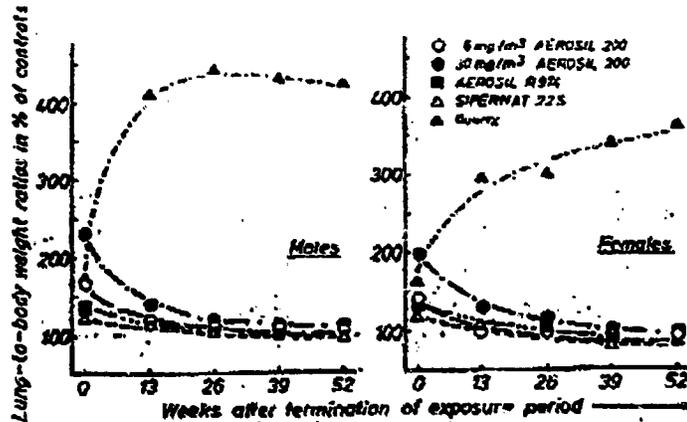


Fig. 1. Relative lung weights expressed as percentage of controls in rats exposed to amorphous silica or quartz for 13 wk and killed at 13-wk intervals after exposure. Exposures were as follows: O, 6 mg Aerosil 200/m<sup>3</sup>; ●, 30 mg Aerosil 200/m<sup>3</sup>; ■, 30 mg Aerosil R 974/m<sup>3</sup>; △, 30 mg Sipernat 22S/m<sup>3</sup>; ▲, 60 mg quartz/m<sup>3</sup>. Values are means for groups of 20 (wk 0 and 52) or 10 (wk 13, 26 and 39) rats.

that could be related to treatment. Urine analyses were essentially negative.

**Organ weights.** At the end of the exposure period both absolute and relative lung weight had increased in all treated groups in comparison with the controls and the increases were statistically significant in all groups except that exposed to 1 mg Aerosil 200/m<sup>3</sup>. The increase was greater in males than in females, and of the groups exposed to amorphous silica, was greatest in that exposed to 30 mg Aerosil 200/m<sup>3</sup> (Fig. 1). During the first 13 wk after exposure, this difference in mean lung weight had decreased considerably or disappeared completely in all groups exposed to amorphous silica. Only in rats exposed to 30 mg Aerosil 200/m<sup>3</sup> did lung weight tend to be higher than in controls as long as 26 wk after exposure. However, lung weight in quartz-exposed animals increased progressively during the post-exposure period to levels three or more times higher than those of the controls. The only other organ that showed increased weight was the thymus in quartz-exposed rats; both the absolute and relative thymus weights were signifi-

cantly increased (by about 140% of control weight in males at day 183). This increase was most pronounced in males and had disappeared 39 wk after the end of exposure.

**Lung collagen contents.** At the end of the exposure period, the lung collagen content of all exposed groups was higher than that of controls (Fig. 2), and the increases were statistically significant in all groups except females exposed to 1 mg Aerosil 200/m<sup>3</sup> or to quartz. The increase was most pronounced in rats exposed to 30 mg Aerosil 200/m<sup>3</sup>. In rats exposed to Aerosil 200 the increase was clearly concentration-dependent and generally more pronounced in males than in females. During the post-exposure period, lung collagen levels gradually decreased in all groups exposed to the amorphous silica, but 1 yr after exposure, only in rats exposed to 1 mg Aerosil 200/m<sup>3</sup> or to 30 mg Sipernat 22S/m<sup>3</sup> lung collagen contents had reached values similar to those of controls. In quartz-exposed rats a completely different pattern was found. At the end of the exposure period, the collagen content was only slightly higher than in

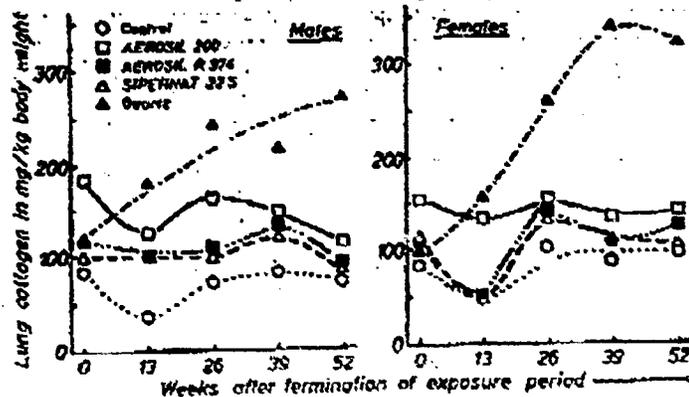


Fig. 2. Lung collagen contents expressed as mg/kg body weight in rats exposed to amorphous silica or quartz for 13 wk and killed at 13-wk intervals after exposure. Exposures were as follows: O, controls; □, 30 mg Aerosil 200/m<sup>3</sup>; ■, 30 mg Aerosil R 974/m<sup>3</sup>; △, 30 mg Sipernat 22S/m<sup>3</sup>; ▲, 60 mg quartz/m<sup>3</sup>. Values are means for groups of 10 (wk 0 and 52) or 5 (wk 13, 26 and 39) rats.

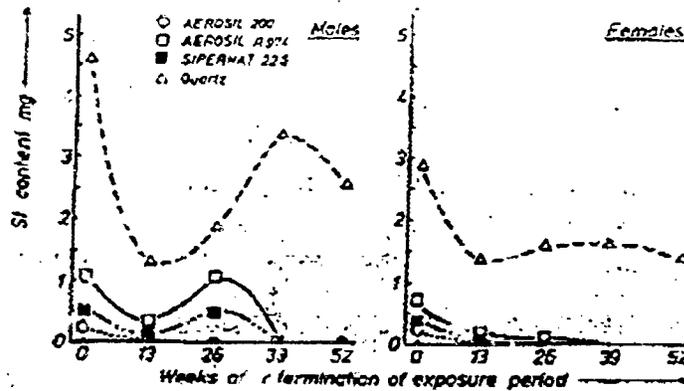


Fig. 3. Total amount of Si in the lungs of rats exposed to amorphous silica or quartz for 13 wk and killed at 13-wk intervals after exposure. Exposures were as follows: ○, 30 mg Aerosil 200/m<sup>3</sup>; □, 30 mg Aerosil R 974/m<sup>3</sup>; ■, 30 mg Sipermat 22S/m<sup>3</sup>; △, 60 mg quartz/m<sup>3</sup>. Values are means for groups of 5 (wk 0 and 52) or 5 (wk 13, 26 and 39) rats.

controls, but in the course of the post-exposure period it had increased markedly.

**Silicon in the lungs and their associated lymph nodes.** Silicon levels in the lungs of males (except those exposed to Aerosil 200) were higher 26 wk after the end of exposure than 13 wk after the end of exposure. Male rats exposed to quartz exhibited even higher values at wk 39 than at wk 26 after exposure. This phenomenon, for which we have no sound explanation, was not observed in females (Fig. 3). In this respect, it should be emphasized that large differences in silicon levels in the lungs were observed between individual animals of the same group at the same stage. Despite the inexplicable results obtained in males the overall picture is clear. Large amounts of silicon were detected in the lungs, and also in the lung-associated lymph nodes of animals exposed to quartz, both at the end of the exposure period and at all stages of the post-exposure period, although silicon levels clearly decreased during the first 13 wk after exposure. Relatively high levels of silicon were found in lungs and associated lymph nodes of rats exposed to Aerosil R 974 at the end of the exposure period, and smaller amounts were present 13 and 26 wk after exposure. Even after a post-exposure period of 52 wk, silicon was present in the lymph nodes of one male exposed to Aerosil R 974. Silicon was detected in the lungs of 100%, and in the lymph nodes of about 50%, of the rats exposed to Sipermat 22S and killed at the end of the exposure period. During the post-exposure period, the amounts of silicon in the lung declined quickly, and 39 wk after the end of exposure no silicon could be detected in the lungs of rats exposed to Sipermat 22S. Sipermat 22S disappeared more slowly from the lung-associated lymph nodes than from the lungs. Aerosil 200 was very quickly cleared from the lungs and the associated lymph nodes.

**Pathology.** Most of the rats exposed to amorphous silicas or quartz and killed at the end of the exposure period had swollen and spotted lungs with a spongy consistency and/or irregular surface and enlarged lung-associated lymph nodes. These changes were most pronounced in the group exposed to quartz. At wk 26 after exposure, the gross changes had

disappeared in all groups exposed to the amorphous silicas. However, in rats exposed to quartz, the gross lesions in the lungs and lung-associated lymph nodes remained present during the whole post-exposure period.

Microscopic changes were mainly observed in the lungs (Table 5). Changes in rats killed at the end of the exposure period comprised slight to severe accumulation of alveolar macrophages, intra-alveolar granular material, cellular debris and polymorphonuclear leukocytes in the alveolar spaces, and increased septal cellularity, seen as an increase in the number of type II pneumocytes and macrophages within the alveolar walls. In general, the most severe changes were found in rats exposed to Aerosil 200 (Plate 2) and quartz (Plate 3), and the mildest changes were in rats exposed to Sipermat 22S. Alveolar bronchiolization, characterized by cuboidal cells lining the alveolar spaces instead of the normal flat cells, occurred mainly in males exposed to 6 or 30 mg Aerosil 200/m<sup>3</sup> or to Aerosil R 974. During the post-exposure periods, no recovery from lung lesions was observed in quartz-exposed rats, whereas in rats exposed to the amorphous silicas the changes disappeared partly or completely. In rats exposed to 30 mg Aerosil 200/m<sup>3</sup> or to quartz accumulations of alveolar macrophages were still found 52 wk after the end of exposure. In rats exposed to Sipermat 22S or Aerosil R 974 these lesions were found until wk 39 after exposure.

Accumulation of intra-alveolar granular material, cellular debris and polymorphonuclear leukocytes were occasionally found in the group exposed to 30 mg Aerosil 200/m<sup>3</sup> and in all quartz-exposed rats during the post-exposure period (Plates 2 and 4). Rats exposed to Sipermat 22S recovered completely from the slight increases in septal cellularity that were observed at the end of the exposure period. A lesser degree of recovery was observed in rats exposed to Aerosil 200 or Aerosil R 974, and no recovery occurred in rats exposed to quartz. Alveolar bronchiolization persisted mainly in quartz-exposed animals and in some rats exposed to Aerosil 200.

Focal interstitial fibrosis, seen as amorphous eosinophilic, collagen-containing thickenings of the septa, was first observed 13 wk after exposure in

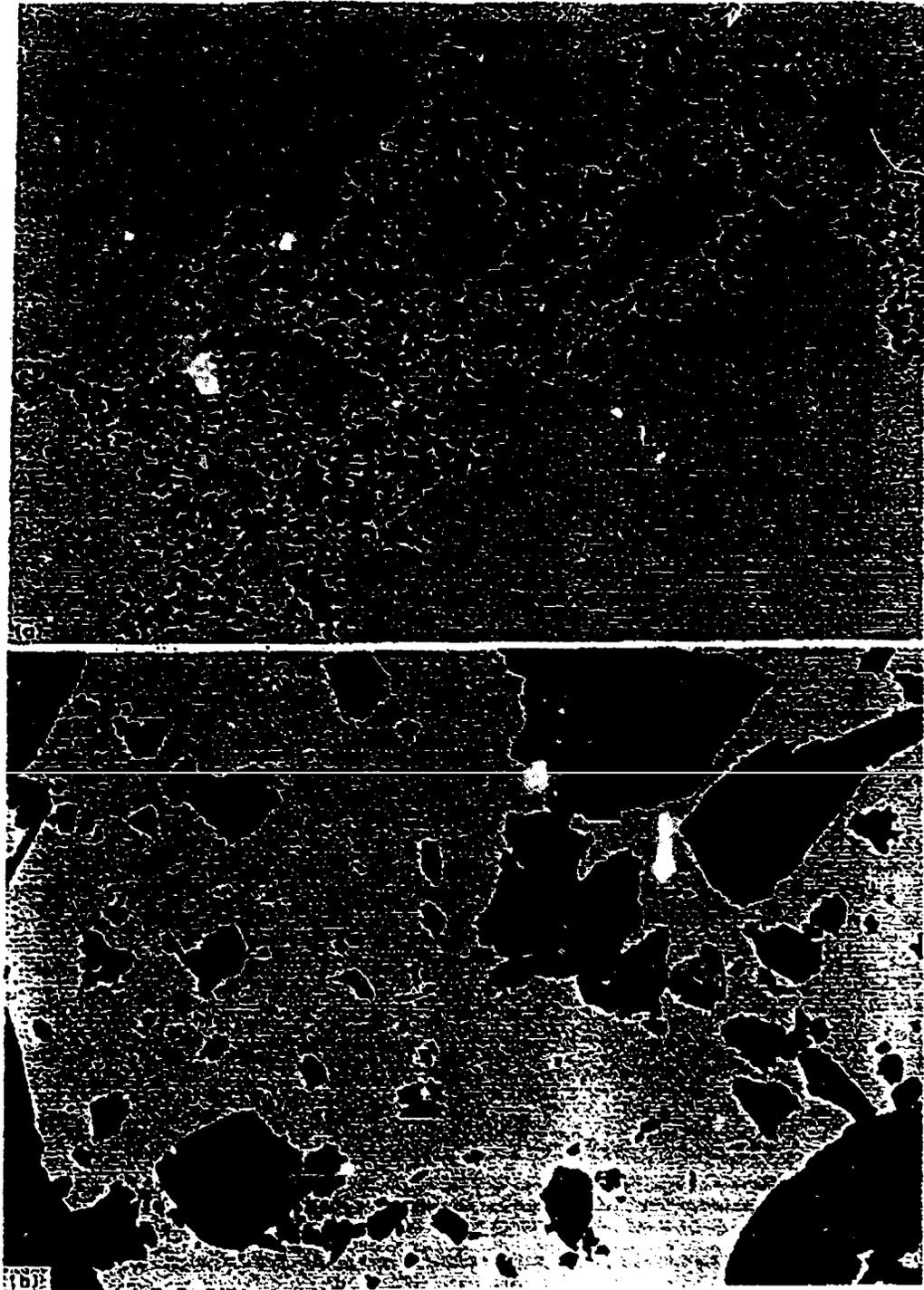


Plate 1. Electron micrographs of (a) an amorphous silica, Aerosil 200 ( $\times 86,400$ ) and (b) a crystalline silica, quartz ( $\times 5760$ ).

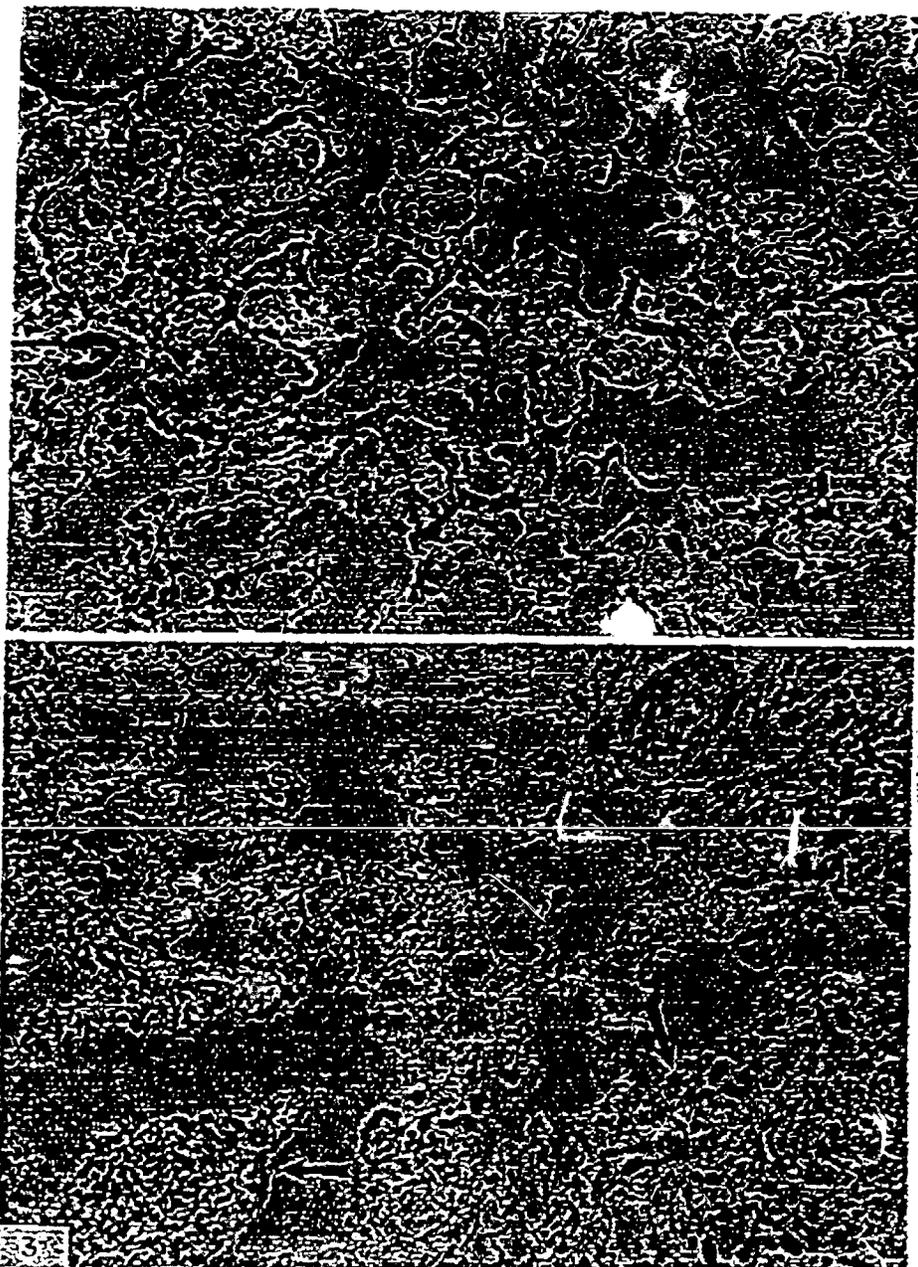


Plate 2. Light micrograph of a portion of the lung from a rat exposed to 30 mg Aerosil 200/m<sup>3</sup> for 13 wk and killed at the end of the exposure period. Note the accumulation of alveolar macrophages accompanied by increased cellularity of the septa. Haematoxylin and eosin. × 160.

Plate 3. Light micrograph of a portion of the lung from a rat exposed to 60 mg quartz/m<sup>3</sup> for 13 wk and killed at the end of the exposure period. Note the granulomatous lesions (arrows) and increased cellularity of the alveolar septa. Haematoxylin and eosin. × 160.

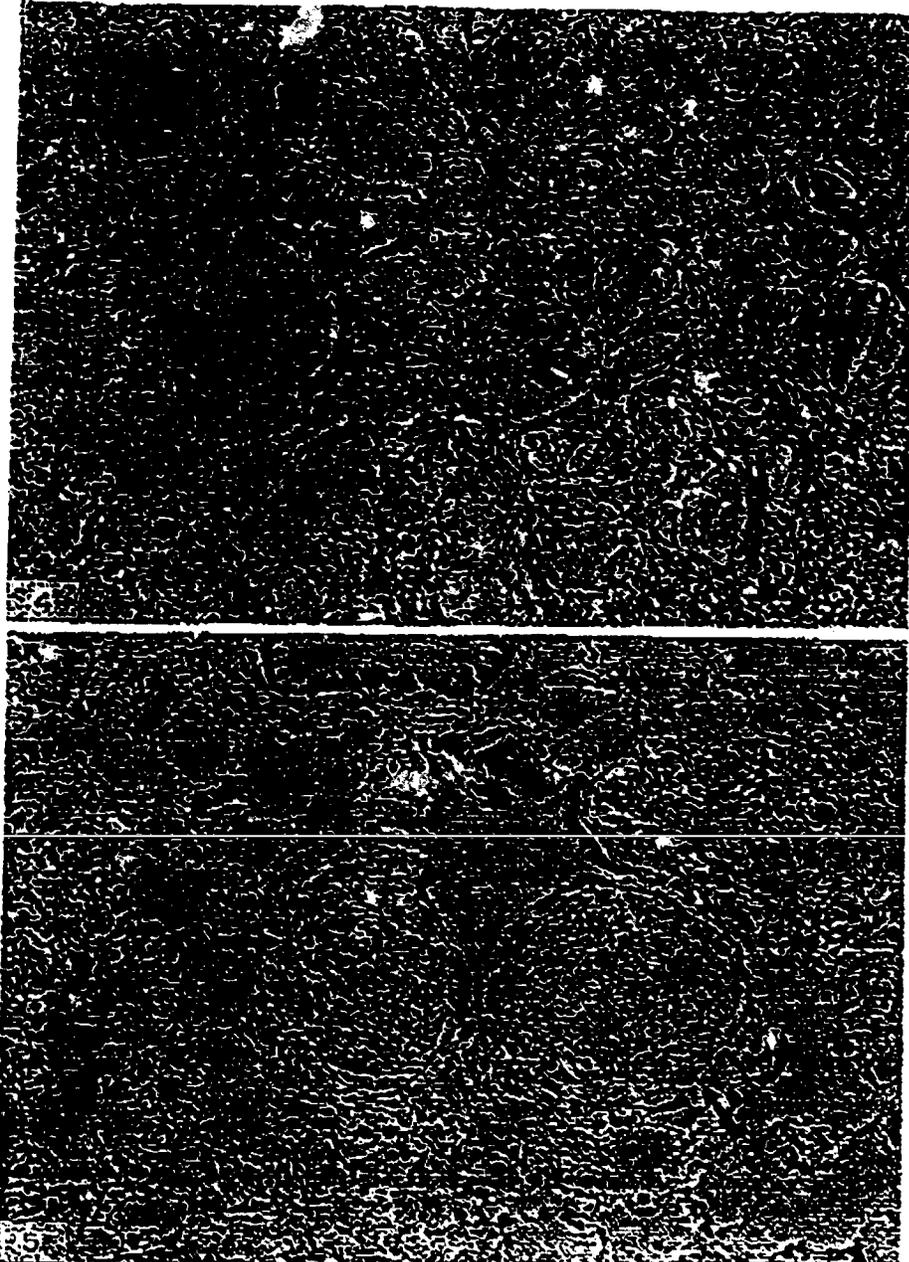


Plate 4. Light micrograph of a portion of the lung from a rat exposed to 60 mg quartz/m<sup>3</sup> for 13 wk and killed after 9 months of recovery. Note the granulomatous lesions comprising accumulations of alveolar macrophages, intra-alveolar accumulation of granular material, cholesterol clefts (arrow) and cellular debris. Haematoxylin and eosin.  $\times 400$ .

Plate 5. Light micrograph of a portion of the lung with granulomas (arrows) from a rat exposed to 30 mg Aerosil R 974/m<sup>3</sup> for 13 wk and killed at the end of the exposure period. Haematoxylin and eosin.  $\times 160$ .

all exposed groups. During the subsequent post-exposure period, this condition disappeared completely in rats exposed to Aerosil R 974 or Sipernat 225, but became more severe in rats exposed to 30 mg Aerosil 200/m<sup>3</sup> or to quartz.

Alveolar cholesterol clefts were seen in some rats exposed to 30 mg Aerosil 200/m<sup>3</sup> 13, 26 and 39 wk after the end of exposure. The presence of these clefts, which are common in chronic inflammatory processes, was associated with the presence of macrophages and polymorphonuclear leucocytes in the alveoli. One male in the 6-mg Aerosil 200/m<sup>3</sup> group also had cholesterol clefts in the lungs 13 wk after the end of exposure. This condition was no longer observed after a post-exposure period of 52 wk. In rats exposed to quartz, cholesterol clefts were observed for the first time after 26 wk of non-exposure. During the remaining recovery period this lesion became more pronounced (Plate 4).

Granulomas, seen as aggregates of macrophage-like cells were scattered throughout the lungs of a few rats exposed to 30 mg Aerosil 200/m<sup>3</sup>, and of all, or nearly all, of those exposed to Aerosil R 974 (Plate 5) or quartz at the end of the exposure period. This lesion disappeared completely in rats of the Aerosil 200 group within 13 wk after the end of exposure, but in rats exposed to Aerosil R 974 recovered only more than 39 wk. Slight fibrosis was demonstrated in the granulomas in animals of the quartz group (Plate 4).

One year after the end of the exposure period, one male rat, which had been exposed to quartz, had a focus of squamous metaplasia in the periphery of the lung. In addition, in one female of the quartz group a small, but unequivocal squamous cell carcinoma was found in the lung parenchyma.

Microscopic changes in the associated lymph nodes were characterized by a considerable accumulation of macrophages with or without cellular necrosis. They were found in all test groups. However, granulomas were not found in the lymph nodes. In the quartz-exposed rats, slight fibrosis of the associated lymph nodes was seen at the end of the post-exposure period.

Treatment-related changes were found in the nose of all rats at the end of the exposure period only. They comprised focal necrosis and rhinitis mainly in animals exposed to the amorphous silica, and slight degeneration of the olfactory epithelium in all exposed groups. The nature of this reversible damage to the nose was similar in all test groups indicating a rather non-specific irritating effect.

The other organs examined, including the liver, did not reveal treatment-related lesions.

#### DISCUSSION

When inhaled, both quartz and the amorphous silicas adversely affected the respiratory tract. Some of the changes were found to be similar for all test materials, but there were also significant differences between the various test materials in the type and degree of the lesions observed. The changes induced by the amorphous silica dusts were generally most apparent by the end of the exposure period, but disappeared more or less quickly within 1 yr after the

end of exposure. The quartz-induced changes were relatively mild at the end of the exposure period but progressed distinctly thereafter, reaching a steady state about 6 months after exposure. There was no recovery during the subsequent 6 months.

The principal effect of quartz is the induction of fibrotic lung disease characterized by an increase in fibrotic tissue and the presence of granulomas (Reiser and Last, 1979; Renne *et al.*, 1985). Amorphous silicas have also been considered to be considerably fibrogenic but, unlike quartz, they have been reported not to induce silicotic nodules (Klosterkötter, 1965). Increases in interstitial fibrosis and granulomatous lesions, and also aggregates of macrophages and polymorphonuclear leucocytes, and septal hypercellularity are considered to be early events in the evolution of silicotic nodules.

In the present study, such early events were seen to different extents in all exposed groups. In rats exposed to the amorphous silicas these lesions regressed, albeit not completely, in rats exposed to Aerosil 200, whereas there was a clear progression of the lesions in rats exposed to quartz. The formation of collagen fibres and hyalinization in the granulomatous lesions, which do not regress, are considered to be late events in the evolution of silicotic nodules. These late events were not found in granulomatous lesions induced by the amorphous silicas. This is in accordance with results of previous studies (Gärtner, 1952; Groth *et al.*, 1979; Klosterkötter, 1965). In quartz-exposed rats, collagen fibres were found in the granulomatous lesions, which did not regress. Therefore, these granulomas were considered silicotic nodules.

The fibrogenic potential of silicon dioxide has been suggested to be attributable to toxic interactions with cell membranes and to the secretion of factors by alveolar macrophages that stimulate lung fibroblast proliferation and collagen formation (Benson *et al.*, 1986; Gritter *et al.*, 1985). In addition, the presence of polymorphonuclear leucocytes in the lungs is considered to play a role in the evolution of lung fibrosis by release of lysosomal enzymes (Adamson and Bowden, 1964). The large numbers of polymorphonuclear leucocytes and alveolar macrophages in the lungs of rats exposed to Aerosil 200 or quartz, and the fibrotic response of the lungs, therefore, are in keeping with this suggestion.

All amorphous silica dusts were completely cleared from the lungs, whereas quartz dust was partially cleared during the first 13 wk after exposure, but not then further cleared. The mechanism by which the test material particles were removed from the lungs in the present study is not fully understood. The role of alveolar macrophages in lung clearance as carriers of phagocytosed particles from the alveoli to the lymph nodes is controversial (Lehnert *et al.*, 1986). However, in the present study accumulations of macrophages laden with fine granular material and the presence of test material in the regional lymph nodes, suggest an active role of the macrophages in lung clearance. Aerosil 200 and quartz dust induced the most pronounced responses of alveolar macrophages, both at the end of the exposure period and during the post-exposure period, despite the fact that of all the materials tested Aerosil 200 was cleared fastest.

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Table 3. Summary of incidences of treatment-related microscopic changes in the lungs of rats exposed to amorphous silica or quartz and then observed for up to 52 wk

Lesion	Time after exposure (wk)	No. of rats	Occurrences of lesion in:														
			Males in Group:						Females in Group:								
			A	B	C	D	E	F	G	A	B	C	D	E	F	G	
Granuloma-like lesions	0	10	0	0	0	0	2	10**	0	10*	0	0	0	0	0	0	0
	13	3	0	0	0	0	1	5**	0	5**	0	0	0	0	0	0	0
	26	3	0	0	0	0	0	3	0	5**	0	0	0	0	0	0	0
	39	3	0	0	0	0	0	1	0	4*	0	0	0	0	0	0	0
	52	10	0	0	0	0	0	0	0	10**	0	0	0	0	0	0	0
Accumulation of alveolar macrophages	0	10	4	10**	10**	10**	10**	10**	10**	10**	1	10**	10**	10**	10**	10**	10**
	13	5	1	2	5**	5**	5**	5**	5**	5**	0	5**	5**	5**	5**	5**	5**
	26	5	0	1	5**	5**	5**	5**	5**	5**	0	5**	5**	5**	5**	5**	5**
	39	5	0	0	3	5**	4*	4*	4*	5**	1	3	4*	4*	4*	4*	4*
	52	10	1	1	10**	10**	3	2	10**	10**	0	7	4	10**	10**	10**	10**
Cellular debris	0	10	0	0	0	0	2	0	0	0	0	0	0	0	0	0	0
	13	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	26	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	39	5	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	52	10	0	0	0	0	1	0	0	10**	0	0	0	0	0	0	0
IPCL	0	10	1	10**	10**	10**	2	2	10**	0	0	0	0	0	0	0	0
	13	5	0	0	0	0	1	0	5**	0	1	3	5**	5**	5**	5**	5**
	26	5	0	0	0	0	0	0	5**	0	0	0	0	0	0	0	0
	39	5	0	0	0	0	0	0	5**	0	0	0	0	0	0	0	0
	52	10	0	0	0	0	0	0	10**	0	0	0	0	0	0	0	0
Increased septal vascularity	0	10	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	13	5	0	0	0	0	0	0	5**	0	0	0	0	0	0	0	0
	26	5	0	0	0	0	0	0	5**	0	0	0	0	0	0	0	0
	39	5	0	0	0	0	0	0	5**	0	0	0	0	0	0	0	0
	52	10	0	0	0	0	0	0	10**	0	0	0	0	0	0	0	0
Alveolar bronchiolization	0	10	1	10**	10**	10**	2	2	10**	1	5**	5**	10**	10**	10**	10**	10**
	13	5	0	0	0	0	0	0	5**	0	1	3	5**	5**	5**	5**	5**
	26	5	0	0	0	0	0	0	5**	0	1	3	5**	5**	5**	5**	5**
	39	5	0	0	0	0	0	0	5**	0	1	3	5**	5**	5**	5**	5**
	52	10	0	0	0	0	0	0	10**	0	0	0	0	0	0	0	0
Focal interstitial fibrosis	0	10	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	13	5	0	0	0	0	0	0	5**	0	0	0	0	0	0	0	0
	26	5	0	0	0	0	0	0	5**	0	0	0	0	0	0	0	0
	39	5	0	0	0	0	0	0	5**	0	0	0	0	0	0	0	0
	52	10	0	0	0	0	0	0	10**	0	0	0	0	0	0	0	0
Cholesterol crystals	0	10	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
	13	5	0	0	0	0	0	0	5**	0	0	0	0	0	0	0	0
	26	5	0	0	0	0	0	0	5**	0	0	0	0	0	0	0	0
	39	5	0	0	0	0	0	0	5**	0	0	0	0	0	0	0	0
	52	10	0	0	0	0	0	0	10**	0	0	0	0	0	0	0	0

19L1 = intra-alveolar polymorphonuclear leukocytic infiltration  
 Circum- were exposed for 13 wk as follows: A—no treatment (control); B—5 mg Aerosil 200/m<sup>3</sup>; C—5 mg Aerosil 200/m<sup>3</sup>; D—30 mg Aerosil 200/m<sup>3</sup>; E—30 mg Aerosil B, 974/m<sup>3</sup>; F—30 mg Silexmet 228/m<sup>3</sup>; G—50 mg quartz/m<sup>3</sup>.  
 Values are for the number of rats shown, and those marked with asterisks differ significantly (Fisher's exact probability test) from the corresponding control value (\*P < 0.05; \*\*P < 0.01).

Sipernat 22S caused only a very slight response of alveolar macrophages, though it was present for more than 26 wk after the end of exposure. In addition to particle size and shape, differences in the chemical and physical properties of the surface of the particles might be responsible for the differences in macrophage response and clearance rate. The dissolution of particles most probably has played the most important role in the lung clearance of the amorphous silicas. The very small primary particles of the amorphous silicas had relative surface areas that were about 10–1000-fold greater than that of quartz. On the basis of this difference in relative surface area the amorphous silica can be expected to have been dissolved considerably faster than the quartz particles. Changes in physico-chemical properties due to chemical treatment of the primary amorphous silica might be responsible for differences in solubility and explain the longer persistence of Aerosil R 974 particles in the lung. The fast dissolution of Aerosil 200 will have resulted in relatively high concentrations of dissolved  $\text{SiO}_2$ , explaining why Aerosil 200 induced more severe lung changes than did Aerosil R 974 or Sipernat 22S.

Mucociliary clearance of the various test materials may have been different, and such possible differences may have been at least partly responsible for the differences in clearance rate between the various test materials.

The toxicological significance of the single squamous cell carcinoma in the lung of one quartz-exposed animal is questionable. However, lung tumours, including squamous cell carcinomas, have been observed in rats both after inhalation (Holland *et al.*, 1986) and after intratracheal instillation of quartz (Groth *et al.*, 1986). In addition, there are indications for the co-carcinogenic properties of quartz (Illing, 1986). Moreover, not one pulmonary squamous cell carcinoma has been found in a total of 1450 Cph:WU, Wistar random rats used as controls in long-term studies carried out in our institute. Therefore, a relationship between the occurrence of the single pulmonary squamous cell carcinoma and the presence of quartz in the lungs is most likely.

The increases in neutrophilic leucocyte counts in rats exposed to Aerosil 200 or quartz were found to parallel the occurrence of changes in the lungs, and indeed were considered to be a reflection of inflammatory pulmonary reactions.

The two most important differences in effects between quartz and the amorphous silicas were as follows:

Quartz, but not the amorphous silicas, induced persistent granulomas very much resembling silicotic nodules.

The adverse effects in the respiratory tract induced by the amorphous silicas partly or completely regressed after exposure was ended, whereas most of the quartz-induced changes progressed.

Of the amorphous silica products, Aerosil 200 generally induced the most severe adverse effects. Despite the very quick clearance of the test material from the lungs and lymph nodes, the recovery of

the respiratory tract occurred rather slowly and incompletely after exposure to levels of 6 or 30  $\text{mg}/\text{m}^3$ . At the 1  $\text{mg}/\text{m}^3$  level the effects were rather mild, and moreover, they disappeared within 13 wk of the end of exposure.

Aerosil R 974 induced effects similar to those of 30  $\text{mg}$  Aerosil 200/ $\text{m}^3$ . The changes were generally slightly less severe and recovery was quicker than in rats exposed to Aerosil 200. The exception was the granulomatous lesions, which occurred more frequently and disappeared at a later stage during the exposure period. This might be related to the higher persistence of Aerosil R 974 in the lungs.

Sipernat 22S induced changes that were also similar to those of both Aerosil products, but they were mild and recovery was quick, even though the test material was only slowly cleared from the lungs and lung-associated lymph nodes.

Quartz at a level of 60  $\text{mg}/\text{m}^3$  induced changes that were similar to those seen in rats exposed to 30  $\text{mg}/\text{m}^3$  Aerosil 200 or Aerosil R 974 at the end of the exposure period, but most changes considerably progressed during the 1-yr post-exposure period. Persistent granulomas, comparable with silicotic nodules, occurred only in lungs of this group. The level of 60  $\text{mg}$  quartz/ $\text{m}^3$  used in the present study was selected on the basis of a 14-day concentration-finding study. Having seen the results of the present study, a lower concentration of quartz (30  $\text{mg}/\text{m}^3$ ) would have been more appropriate because it would have allowed an even better comparison of the effects of quartz with those of the amorphous silicas.

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