GENERAL REMARKS ON THE SUBSTANCES CONSIDERED

This sixty-eighth volume of *IARC Monographs* considers certain forms of crystalline and amorphous silica, some silicates (palygorskite, also called attapulgite; sepiolite; wollastonite; and some natural and synthetic zeolites, excluding erionite), coal dust and *para*-aramid fibrils. Some of these agents are fibrous in nature (palygorskite, sepiolite, wollastonite and some natural zeolites, as well as *para*-aramid fibrils). With the exception of coal dust, zeolites (other than erionite) and *para*-aramid fibrils, these agents were evaluated by previous IARC working groups in 1986 (IARC, 1987a,b) (see **Table 1**). Since these previous evaluations, new data have become available, and the Preamble to the *IARC Monographs* has been modified (Vainio *et al.*, 1992) to permit more explicit inclusion of mechanistic considerations and of data on aspects other than cancer in the evaluation process.

Table 1. Previous evaluations[a] of agents considered in this volume

Agent	Degree of carcinogenicity		Overall evaluation of carcinogenicity to humans
	Human	Animal	
Silica, crystalline	L	S	2A
Silica, amorphous	I	I	3
Wollastonite	I	L	3
Attapulgite (palygorskite)	I	L	3
Sepiolite	I	I	3

S, sufficient evidence; L, limited evidence; I, inadequate evidence; Group 2A, probably carcinogenic to humans; Group 3, cannot be classified as to its carcinogenicity to humans (see also Preamble, pp. 23–27)

[a] *IARC Monographs* Volume 42 (IARC, 1987a) and Supplement 7 (IARC, 1987b)

Factors affecting toxicity of inhaled materials

Physical and chemical properties may play an important role in the degree of exposure and subsequent toxicity of inhaled materials. Properties such as chemical composition, particle diameter, particle surface area, shape, density, solubility, and hygroscopic and electrostatic properties may be important factors that affect toxicity resulting from inhalation of particles.

The durability or biopersistence of particles or fibres can be defined as their retention in the lung over time. Important parameters that may be altered by residence in the lung are particle or fibre number, dimensions, surface reactivity, chemical composition and surface area. Particulates can be eliminated from the lung by mechanical clearance, primarily involving macrophage uptake and transport to the mucociliary escalator, or by dissolution. The biopersistence of particulates in the lung is dependent upon the site and rate of deposition, as well as rates of translocation, clearance, dissolution and biomodification of the particulate in the lung. The clearance, as well as toxicity, of particulates deposited in the respiratory tract is influenced by the solubility of the particulates in water and tissue.

Surface-related factors which have been postulated to influence particulate-induced toxicity and carcinogenicity include (1) the presence of iron or other transition metals; (2) the ability of a particle to accumulate iron; (3) the ability of particulates to generate free radicals; and (4) hydrophobicity of the particulate surface.

Complexities in assessing exposures to mineral dusts

Since human exposures occur via inhalation of solid particulates, it is useful to consider just some of the ways in which particles are characterized. Particles are frequently described in various size ranges, including coarse, fine and ultrafine. Coarse particles are typically described as those with a diameter > 2 μm; fine particles are those in the range of 0.1–2.0 μm; and ultrafine particles are described as those with a diameter < 0.1 μm. While particle size is often characterized as the geometric mean diameter in inhalation studies, the aerodynamic characteristics of the particles are of importance.

It is important to bear in mind that in humans inhalation of 'respirable' particles entails exposures to those particles in a mineral dust that are able to penetrate into the alveolar spaces of the lungs. It is generally considered that particles with an aerodynamic diameter of less than 3–4 μm are respirable, while most particles greater than 5 μm may not reach the alveolar region because of their deposition in the tracheobronchial airways.

Particle shape is also known to play an important role in influencing the pathogenesis of particle-associated lung disease. This has been especially well demonstrated in the case of fibres. Fibres are defined by length : diameter ratio (aspect ratio) with lengths being at least three times the diameter. Fibre diameter generally determines the respirability of the sample and fibre length strongly influences its biological activity.

Minerals rarely occur in a pure form in nature. At some sites within a crystalline structure, one element may be substituted for another. Minerals also occur in a range of forms and morphological habits and with other minerals. Such variations affect the biological activity of minerals and powdered admixtures. For example, silica polymorphs, including quartz and its varieties, can contain trace impurities that affect the biological activity of 'free silica'. Wollastonite, derived by metamorphism of dolomite rocks, can not only vary chemically but can also occur geologically with fibrous amphiboles. Palygorskite and sepiolite clays vary considerably with regard to chemistry, crystal form, fibre length and the presence of associated materials. *para*-Aramid fibrils are formed from the peeling of fibres under conditions of abrasion and their physical

characteristics may vary depending on the conditions under which they are generated. Coal dust is a complex and variable mixture. Exposure to coal dust occurs mainly in coal mines where there are also exposures to other agents, e.g. diesel exhaust and silica.

Occupational exposures to mineral dusts are therefore particularly complex. A mineral mixture to which workers are exposed may differ according to geological source. Workers in different processes, such as mining and milling, production and use, may be exposed to different mineral varieties, especially if extensive beneficiation is employed; or they may be exposed to single minerals with very different properties, such as particle size, surface properties and crystallinity, due to alterations during industrial processing.

Problems encountered in the evaluation of epidemiological studies

The available epidemiological information on cancer risks associated with crystalline silica is solely based on findings from occupationally exposed populations. Only sporadic data on environmental exposure were available and were therefore not considered in the epidemiological assessment. Although there is a relatively large body of epidemiological data, there are some important areas of uncertainty that complicate the epidemiological assessment. Some of these uncertainties relate to the inherent difficulties encountered in studying occupational populations for cancer risk. These include limitations in the amount and quality of historical exposure data relevant to cancer induction times; deficiencies in data on potentially confounding factors, such as exposure to radon or cigarette smoking; and difficulties in the interpretation of chest radiographs as evidence of exposure. The most severe of these limitations is the generally absent or minimal data on occupational hygiene measurements to enable exposure–response estimation for crystalline silica. However, the Working Group's evaluation of the epidemiological evidence for potential causal relation between silica and cancer risk was focused principally on findings from studies that were likely to have been distorted by confounding and selection biases. Among these studies, those that addressed exposure–response associations were especially influential in the Working Group's deliberations.

Problems encountered in the evaluation of experimental studies

Hazards associated with inhalation of particulate materials including fibres require toxicological considerations which are different from those needed for other substances. It is generally regarded that physical dimensions, durability or biopersistence, and surface characteristics are important factors in the production of particle-related pathological effects in the lungs of exposed humans and experimental animals. The following discussion reflects, in part, the concepts developed in IARC Scientific Publication No. 140 on Mechanisms of Fibre Carcinogenesis (Kane *et al.*, 1996). Some of these considerations also apply to other fibrous materials previously evaluated in *IARC Monographs* but not reviewed in this volume, including asbestos (IARC, 1977), man-made mineral fibres (IARC, 1988) and the naturally occurring zeolite, erionite (IARC, 1987a).

Chronic inhalation studies in rats have demonstrated that numerous kinds of particles, when inhaled at various concentrations, can induce significant adverse effects, including impaired pulmonary clearance, prolonged lung inflammation, pulmonary fibrosis and lung tumours. These effects have been observed in the lungs of rats following inhalation of highly cytotoxic materials such as crystalline silica, as well as with particles of other substances of low solubility and low cytotoxicity (e.g., talc, titanium dioxide). Concentrations of inhaled particles have ranged from as low as 1 mg/m^3 for quartz to 250 mg/m^3 for titanium dioxide. Other particulate materials which have been investigated include diesel exhaust, coal dust and carbon black. Lung tumour incidences in chronically exposed rats have ranged from 3 to 40%, depending upon the material, different particle sizes and concentrations. These findings may be accounted for, in part, by the deposition efficiencies of the inhaled particles in the lung, different particle sizes and particle surface areas and/or the cytotoxicity/reactivity of the inhaled dusts. It is important to note that the development of particle-induced lung tumours occurs in rats, but not to any great degree in mice or hamsters. Clearly, a difference exists in the pulmonary responses of rodent species to chronic exposures to inhaled dusts.

The mechanisms underlying the rat lung response have not been fully elucidated. The results of a number of studies suggest that there may be common mechanisms for induction of rat lung tumours observed in response to chronic inhalation of low-solubility particles. Tumours arise in lungs in which there is significant chronic inflammation, epithelial hyperplasia and metaplasia and parenchymal pulmonary fibrosis. In this respect, there is increasing evidence supporting the hypothesis that the tumours represent a generic response of the rat lung to particle-elicited persistent pulmonary inflammation and increased epithelial cell proliferation. In this mechanism of induction of rat lung tumours by particles, inflammation and the associated release of cell-derived oxidants are hypothesized to produce a genotoxic effect, while enhanced epithelial cell proliferation increases the likelihood that any oxidant-induced or spontaneously occurring genetic damage becomes fixed in a dividing cell and is clonally expanded. Thus, it is postulated that when a 'threshold' particle dose is exceeded chronically in the rat lung there develops an inflammatory and cell proliferative response sufficient to increase the probability of genetic changes necessary for neoplastic transformation to occur.

Certain physical characteristics may have special relevance for fibre toxicity. One example is the parameter of fibre dimensions. Fibre dimensions, which involve both diameter and length parameters, are known to play an important role in influencing the pathogenesis of fibre-associated lung disease. This has been demonstrated clearly by Davis *et al.* (1986) who carried out a one-year inhalation study with rats exposed to aerosols of specially prepared 'short' (i.e. < 5 μm in length) amosite asbestos fibres or to a preparation of long (i.e. > 20 μm in length) amosite asbestos fibres, both preparations derived from the original source and at equivalent gravimetric concentrations. Thus, rats were exposed to greater numbers of short amosite fibres than long amosite fibres. Following the one-year exposure, no histopathological effects were observed in rats exposed to the short fibre preparation, while one-third of the rats exposed to gravimetrically similar concentrations of long amosite fibres developed lung tumours. In addition, nearly all of the rats exposed to the long fibres concurrently developed diffuse

pulmonary fibrosis. Similar dimension-related differences have been reported by Davis and Jones (1988) in studies of chrysotile asbestos in rats. Gilmour *et al.* (1995) demonstrated enhanced free radical activity of long amosite fibres when compared to short amosite fibres. The interpretation of animal inhalation studies of particulate materials thus clearly requires careful characterization of the physical dimensions of the particles as well as their surface reactivity.

A final complexity in extrapolating from experimental studies in animals to human experience is that there are virtually no studies in which exactly the same material to which humans are exposed has been systematically evaluated in experimental animals.

Relevance of in-vitro assays

At present, there is insufficient understanding of how the physical and chemical properties of fibres contribute to mechanisms of fibre-induced carcinogenesis. However, there are physical and chemical properties of fibres that have been associated with fibre toxicity *in vitro* and toxicity and/or carcinogenicity *in vivo*, particularly free radical generation. In contrast, the results of cytotoxicity tests with fibres *in vitro* appear to be dependent on fibre length (Hart *et al.*, 1994). In-vitro studies with non-fibrous particles may correlate better with in-vivo effects. Nevertheless, characterizing selected physical and chemical properties of particulates could be useful in the context of screening assays to make inferences on the relative potential of fibres to produce adverse effects *in vivo*. However, given the current limitations of in-vitro particulate testing, these inferences require validation using in-vivo experiments.

Relevance of short-term in-vivo assays

As discussed above, experimental studies with particulates in rats demonstrate a correlation between significant numbers of lung tumours and high levels of pulmonary fibrosis. Particulate-induced chronic inflammation leads to fibrosis and is frequently associated with increased levels of epithelial hyperplasia, as demonstrated by increased epithelial cell proliferation. Chronic inflammation and hyperplasia have also been associated with the development of lung tumours, particularly in rats. Short-term in-vivo assays may have value in predicting particulate-related, long-term pathological effects, including lung tumours.

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