Health Hazard Assessment of Nonasbestos Fibers

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EXECUTIVE SUMMARY

The inhalation of asbestos fibers including amosite, chrysotile, and crocidolite has been associated with the development of lung cancer, mesothelioma, pulmonary fibrosis and other nonmalignant pleural diseases in humans. Because the pathogenic effects of asbestos are attributed, in general, to its fibrous nature, human health concern extends to the use of other fibrous substances. This document assesses the health effects of nine non-asbestos fibers and attempts to determine the nature and magnitude of the health hazard as compared to asbestos. The fibers chosen for investigation were selected for one or more of the following reasons: a) they are commercially important; b) they are considered potential asbestos substitutes; c) they represent fiber types with broadly different physical and chemical characteristics; and, d) health data are available on them. The fibers evaluated in this report include fibrous glass, mineral wool, ceramic fibers, erionite, wollastonite, attapulgite, aramid fibers, carbon fibers, and polyolefin fibers.

Available data suggest some similarities in the health effects of asbestos and some nonasbestos fibers but the degree of the health effects may differ substantially among fiber types. The differences in the biological activity may be associated with the specific characteristics of each fiber type including fiber morphology, size distribution, chemical constitution, surface properties and durability.

A basic property which allows a fiber's potential toxicity to be expressed is its respirability, i.e., its ability to penetrate into the smaller conducting airways of the tracheobronchilar tree

and the alveolar region of the lung. It is clear that fiber diameter is the most important factor in determinig the respirability of the fiber. Fiber length and morphology also affect the respirability of the fiber but to a lesser extent. Also, it would appear that the fiber needs to be retained and persist in the tissue in order to cause toxicity. Fiber length is an important determinant of fiber retention, with shorter fibers being cleared more readily. Fiber retention is also determined by the biological solubility of fibers which is directly related to their chemical composition and physical characteristics.

To date, the exact role of various fiber properties in relation to biological activity and pathogenicity is not clearly understood. It is clear, however, that different types of fibers with similar size properties (e.g. erionite and asbestos) could have very different biological activity, although there is increasing evidence suggesting that for a given fiber type, fiber size is an important factor, i.e., the thinner and longer the fiber, the more hazardous it is. Additional research is necessary to examine further the importance of fiber properties in mediating the induction of disease and investigate the mechanisms by which fibrous materials cause disease.

It is difficult to definitively assess the relative biological activity and pathogenicity of nonasbestos fibers in comparison to asbestos because of limited data bases. Major limitations include a lack of comparable dose-response data as well as information available on the characteristics of the tested fibers, particularly, fiber morphology and size

distribution and the number of fibers in each size category. However, on the basis of available information, it may be concluded at this time that with the possible exception of erionite, the other eight fibers reviewed in this report do not pose a health hazard of similar magnitude as asbestos. Additional studies are needed to conclusively determine the health effects of each fiber type. Erionite, which may be more hazardous than asbestos, is not a major concern because of its limited production and use.

A summary of the hazard assessment of the oncogenic and fibrogenic effects of these fibers, and the testing recommendation(s) to fill data gaps for each fiber is presented in the following sections. The assessment of the potential for carcinogenicity of fibers in humans is based on the current U.S. EPA classification system for categorizing the overall weight-of-evidence as determined from human, animal and other supporting data (USEPA, 1986).

Inhalation is the major route of exposure to fibers and exposure via this route of administration has been shown to cause cancer in humans in the case with asbestos. Hence, it would seem most relevant to use the inhalation route for the animal testing of fiber carcinogenicity. Positive results from inhalation studies in animals would be interpreted to have significant implications for potential hazard in the human since asbestos has also been found to induce tumors in animals following inhalation exposure. The major pathogenic effects associated with the inhalation of asbestos in humans including lung fibrosis, lung cancer and mesothelioma have been replicated in rodents exposed

to asbestos by inhalation. There are, however, shortcomings of inhalation studies. One reason is that fiber deposition and retention in rodents are considerably different from those in humans. Rodents, being obligatory nose breathers, have a greater filtering capacity than humans resulting in a lower alveolar deposition of fibers in rodents. As a result, inhalation tests in rodents may underestimate the hazard potential of fibers to humans unless it is clear that the number of fibers reaching the target tissues are comparable to the positive control.

Experimental procedures other than inhalation exposure testing have been developed which attempt to accommodate for these species differences and to achieve comparable target organ doses. However, they do have their disadvantages. In these studies, the test fiber is artificially introduced in large "bolus" dose(s) directly into the target tissue such as the mesothelium as in the cases with intraperitoneal and intrapleural administrations, or near major targets including the lung and pleural mesothelium in the case of intratracheal instillation. Caution must be excercised in extrapolating the findings from parental administration studies in animals to humans, since the results from such studies may not be predictive of inhalational hazard. Injection studies bypass the normal physiological deposition and clearance mechanisms and lead to non-random accumulations of test substances at the site of deposition. Thus, respirability characteristics, which are routinely taken into account in an inhalation study, are not operative following injection. Nevertheless, injection studies are of value by providing useful information regarding the intrinsic biological

activity of the test fiber under conditions where the material is in direct contact with the cells at risk.

1. Fibrous Glass

There is no evidence in available epidemiologic studies that peritoneal or pleural mesotheliomas are associated with occupational exposure to man-made mineral fibers (fibrous glass and mineral wool). With regard to the respiratory cancer risk, there was no excess of such cancers among continuous glass filament workers in either the U.S. or Europe. For glass wool production workers, there was no significant increase in mortality from respiratory cancer (or lung cancer) compared with regional rates in either the U.S. or European cohorts, though there were statistically significant small increases compared with national rates in the U.S. study. In both investigations, mortality from respiratory cancer increased nonsignificantly with time from first exposure. However, it was not related to the duration of employment or cumulative fiber exposure in the U.S. study. Also, in the European study, it was not related to the duration of exposure or to different technological phases reflecting differences in the intensity and quality of exposure. A lack of dose-related trends might be due in part to the very low exposure experienced by the cohorts.

Among glass wool workers in the U.S. cohort who were ever exposed to small diameter fibers (<3.0 μm), there was a nonsignificant excess of respiratory cancer mortality which increased nonsignificantly with time since first exposure, compared to those who had never been exposed. A third study

reported a statistically significant increase of lung cancer mortality among Canadian glass wool workers, but this was not related to the time since first exposure nor to the duration of exposure.

On the basis of available information, the evidence for carcinogenicity of small diameter glass fibers, glass wool and glass filament from studies in humans is considered inadequate. Still, the epidemiologic findings seem to suggest that workers engaged in the manufacture of glass wool and small diameter fibers might be at increased risk of developing respiratory cancer; additional studies are necessary to clarify the health effects of fibrous glass in humans.

A number of long-term inhalation studies have not provided evidence of lung tumor or mesothelioma in several animal species exposed to glasswool (typically 3-10 μm in diameter), fine fiber-glass (1-3 μm in diameter) or to very fine fibrous glass (also known as glass microfibers; <1 μm in diameter). Shortcomings of these investigations include the use of small numbers of animals, relatively short fibers, low numerical concentrations of fibers, limited study duration, and/or inadequate positive control. In contrast to the inhalation studies, many animal studies involving the intrapleural injection/implantation, or intraperitoneal injection of fine glass fibers or glass microfibers consistently demonstrate that these fibers are capable of producing mesothelioma in rats, hamsters, and mice when they are introduced directly into the body cavity. Glass wool has also been shown to produce low incidences of pleural tumors in a few intrapleural implantation/injection studies in rats. In addition, an

increased incidence of both lung tumors and pleural mesothelioma has been reported in one study following intratracheal administration of glass microfibers to hamsters; this indicates that under certain conditions, glass fibers can pass through the lung and incite reactions in the pleura. In another intratracheal instillation study, glass microfibers also caused lung tumors in the rat. However, several other intratracheal instillation studies in hamsters and rats have not reported tumor formation with glass wool, fine glass fibers or glass microfibers.

In the absence of positive findings from available inhalation studies, the evidence for human carcinogenicity of very fine and fine fibrous glass and glass wool from animal studies is considered limited because only non-physiological routes of administration are associated with carcinogenic findings. However, the repeated observation of tumors following these administrations do indicate the biological activity of the test fibers when deposited in high enough quantity at or near the target tissue. The animal data are supported by positive findings from a few genotoxicity studies which indicate that fine fiberglass and glass microfibers cause similar weak genotoxic effects (clastogenicity and cell transformation) generally seen with asbestos. Thus, considering all available data (human, animal and supporting evidence), the Office of Toxic Substances (OTS) of the U.S. Environmental Protection Agency (USEPA) proposes to classify fine and very fine fibrous glass and glass wool in Category C, i.e., possible human carcinogen, mainly based on inadequate evidence of carcinogenicity in humans and

limited evidence in animals. Others might interpret the same information as supporting a B2 (probable human carcinogen) designation, mainly because they think the animal injection studies should be afforded more weight. Irrespective of these differences in classification, all would agree that the existing evidence supporting a human carcinogen hazard for fibrous glass is much less convincing than for asbestos.

As for the continuous glass filament (nominal diameters of 6-15 μm), there is inadequate evidence of carcinogenicity in laboratory animals. The results of a few available intrapleural implantation studies showed that large diameter glass fibers did not induce mesothelioma in rats. Glass filament is therefore not classifiable as to human carcinogenicity on the basis of inadequate evidence of carcinogenicity in humans and animals (Category D).

There does not appear to be any convincing evidence for increased risks of non-malignant respiratory disease (NMRD) among fibrous glass workers. In the European study, there was no excess mortality from NMRD in the continuous glass filament or glass wool cohort, nor was there a trend with time since first exposure or duration of employment. Similarly, in the U.S. study, no significant excess of NMRD mortality was found among glass filament workers compared with either local or national rates. For U.S. glass wool workers, there was no significant increase in NMRD mortality based on local rates though there was a statistically significant excess compared with national rates. Further analyses of NMRD mortality showed no apparent dose-related trends. Among glass wool workers ever exposed to

small diameter fibers, no excess of NMRD mortality was observed but there was a nonsignificant increase with time since first exposure. The results of a respiratory morbidity study showed some evidence of radiographic opacities in the lung of a limited number of fibrous glass workers but there was no evidence of pulmonary fibrosis, no increase in respiratory symptoms and no impaired lung function.

Long-term inhalation studies have not provided definitive evidence for the development of lung fibrosis in laboratory animals exposed to fine glass fibers or glass wool. However, the positive findings from several injection studies in animals and in vitro cytotoxicity studies indicate that fine and very fine fiberglass may be fibrogenic.

Overall, it may be concluded that a possible health hazard exists from inhalation exposure to fine and very fine fibrous glass, i.e., fibers with diameters consistently below 3 microns. A low health concern is also raised for exposure to glass wool which does contain some respirable fine fibers. As for continous glass filaments which are generally nonrespirable, they would appear to pose little or no hazard to exposed humans. On the basis of available experimental data, it is concluded that fibrous glass appears to be less pathogenic than asbestos. Although the fibrogenicity and oncogenicity of fine fibrous glass and glass wool have been extensively investigated, none of the available inhalation studies are considered adequate. Furthermore, since considerable data gaps still exist, particularly a lack of comparative dose-response effects with asbestos, additional inhalation/injection studies would be

useful. It also appears necessary to conduct additional epidemiological studies to conclusively determine the health hazard potential of fibrous glass in humans.

2. Mineral Wool

Small excesses of mortality due to respiratory cancer have been observed among rock wool/slag wool workers in the U.S. and in Europe. In the U.S. study, the excess of respiratory cancer mortality was statistically significant when compared to both local and national rates. There was no clear trend with time since first exposure and there was no relationship with duration of exposure, cumulative fiber exposure or average intensity of exposure. The results of a nested case-control study using cases from the U.S. cohort showed a weak but positive trend between mineral wool exposure and respiratory cancer when confounding by cigarette smoking was considered.

In the European study, the lung cancer excess found among rock wool/slag wool workers was not statistically significant compared with either local or national mortality rates. There was also a statistically nonsignificant increased mortality with time since first exposure but there was no relationship between lung cancer mortality and duration of exposure. The highest and statistically significant lung cancer rates were found among workers after more than 20 years first exposed in the early technological phases, during which fiber airborne levels were presumably higher than in later production phases. The presence of workplace contaminants such as bitumen, pitch or asbestos could not explain the observed lung cancer excess.

Overall, the available epidemiological findings suggest that mineral wool workers are at increased risk of respiratory cancer. The evidence for mineral wool as an etiological agent includes the consistent elevated risk observed in several rock wool/slag wool facilities, and the higher cancer risks found among workers who had twenty or more years elapse since first exposure. The evidence not supporting an etiological relationship is the lack of a consistent dose-response trend. This might be due in part to the low levels of fiber exposure and the potential exposure misclassification. On the basis of available information, the evidence for carcinogenicity of mineral wool from epidemiological studies is considered limited.

The results of three limited long-term studies showed that mineral wool did not produce tumors in rats or hamsters when administered by inhalation. However, mineral wool has been shown in a few studies to induce varying tumor yields in rats via either the intrapleural (pleural mesothelioma) or intraperitoneal route (peritoneal mesothelioma) of exposure. Overall, the experimental evidence for the carcinogenic potential of mineral wool is considered to be limited. Thus, OTS is proposing to classify mineral wool as a probable human carcinogen (Category B1) on the basis of limited evidence of carcinogenicity from epidemiological studies and limited evidence from animal studies. There is no genotoxicity information available on mineral wool.

There is inadequate epidemiological evidence for an association between the development of non-malignant respiratory diseases (NMRD) and exposure to mineral wool. No increased

mortality from NMRD was found for the European rock wool/slag wool workers. In the U.S. study, a statistically nonsignificant excess of NMRD mortality was observed among mineral wool workers based on local or national rates. However, there was no relationship with time since first exposure, duration of exposure, average intensity of exposure, or estimated cumulative level of exposure. Furthermore, the results of a respiratory morbidity study in the U.S. showed no evidence for impaired lung functions or radiographic lung abnormalities associated with mineral wool exposure.

There is little experimental evidence for the fibrogenicity of mineral wool. Mineral wool was not found to cause lung fibrosis in three long-term inhalation studies but focal fibrosis was reported in a very limited inhalation study involving only two rats. The results of two in vitro studies showed that mineral wool was cytotoxic in cells in culture. In view of these findings, concerns for possible development of pulmonary fibrosis associated with mineral wool exposure cannot be entirely ruled out at this time.

Based on the limited data base in animals, mineral wool appears to be less biologically active and less pathogenic than asbestos fibers. It is concluded at this time that mineral wool fibers may present a health hazard to exposed humans but not to the same magnitude as asbestos. Since the pathogenic effects of mineral wool have not been adequately characterized, additional epidemiological studies and animal testing are needed.

3. Ceramic Fibers

There are no studies available on the potential health effects from exposure to ceramic fibers in humans. The pathogenicity of ceramic fibers in laboratory animals appears to vary considerably for different fiber types which may be a function of variation in fiber size distribution.

An increased incidence of lung tumors have been observed after chronic inhalation exposure to ceramic aluminum silicate glass in one study using rats. Another inhalation study produced no tumors in rats, but one mesothelioma in a hamster. An intratracheal instillation study conducted by the same laboratory showed no tumor induction with refractory aluminum silicate fibers. However, these fibers have been shown in several long-term studies to cause mesothelioma in rats and hamsters by intrapleural or intraperitoneal injection. Based on the sufficient evidence of carcinogenicity in animals in multiple experiments with different routes of administration, but in the absence of human data, OTS proposes to classify ceramic aluminum silicate fiber as a probable human carcinogen (Category B2).

The experimental evidence of fibrogenicity of ceramic aluminum silicate fibers is limited. The positive results of a chronic inhalation study suggest that long-term inhalation of ceramic aluminum silicate glass may produce mild interstitial lung fibrosis in humans. This finding is further supported by positive findings from an *in vitro* cytotoxicity study of ceramic aluminum silicate glass.

In view of available findings and since ceramic aluminum silicate fibers are respirable and durable, it may be concluded that this ceramic fiber type may present a health hazard to the exposed humans. Because of the variable results from available *in vivo* and *in vitro* studies on ceramic aluminum silicate, its relative pathogenicity in comparison to asbestos cannot yet be made at this time. In order to further evaluate the health effects of ceramic aluminum silicate fibers, it is recommended that epidemiological studies of exposed workers be initiated. No additional animal tests are recommended at this time since a large-scale animal study by various routes of exposure is currently being conducted at a private laboratory.

Available animal studies have not provided evidence of the carcinogenicity and fibrogenicity for refractory alumina oxide and zirconia oxide fibers. It has been shown in several studies that these fibers did not produce tumors nor fibrosis in rats via chronic inhalation exposure or by intracavitary injection. The lack of experimental pathogenic effects of these fibers may be attributable to the test fibers being largely nonrespirable. Similarly, the cytotoxicity of these fibers in rat peritoneal macrophages is low. These refractory fibers are therefore not classifiable as to human carcinogenicity (Category D) on the basis of inadequate evidence of carcinogenicity in animal studies and in the absence of human data. Based on available findings, it would appear that refractory alumina and zirconia fibrous products containing mostly nonrespirable fibers would not pose significant health hazard in exposed humans.

4. Erionite

Available epidemiological data show that populations from South Central Turkey have an excessive incidence of malignant pleural mesothelioma and nonmalignant pleural diseases (chronic pleurisy fibrosis, pleural thickening and pleural plaques). The etiology of these diseases is uncertain but there is limited evidence to indicate that erionite fibers may be the major etiological factor. All of the experimental studies conducted to date have confirmed that erionite from Turkey and deposits in the U.S. causes a significant increase in malignant meseothelioma in animals by several routes of exposure including inhalation. Animal data are also supported by findings that erionite is genotoxic, in causing DNA damage and repair and inducing cell transformation in culture. Thus, OTS is proposing to classify erionite as a probable human carcinogen (Category B1) on the basis of limited evidence of carcinogenicity from studies in humans and sufficient evidence of carcinogenicity from animal studies.

There is no information available on the ability of erionite to induce fibrotic diseases in animals by inhalation. However, erionite has been shown to cause fibrogenic effects in animals by the injection method. Furthermore, available in vitro studies demonstrate that erionite is hemolytic and highly cytotoxic. Thus, it is concluded that erionite is potentially fibrogenic in view of the limited evidence from epidemiological studies and limited evidence from experimental studies.

Overall, there is sufficient evidence to conclude that erionite potentially poses a significant health hazard to exposed

humans. However, if practical, additional epidemiological studies should be conducted to further evaluate the association between erionite environmental exposure and development of malignant and nonmalignant respiratory diseases. Since the toxicological profile of erionite has been adequately characterized in animals, no further testing is recommended. Based on the available experimental data, erionite appears to be at least as hazardous as asbestos.

5. Wollastonite

None of the available epidemiological studies were designed to assess the risk of lung cancer or mesothelioma associated with wollastonite exposure. One case of mesothelioma has been reported in a worker who had been exposed to wollastonite, but no cause and effect relationship can be drawn based on a single case report. Preliminary information on an inhalation oncogenicity study of wollastonite in rats indicates the lack of a tumorigenic response. The results of an intrapleural implantation study showed that wollastonite was weakly tumorigenic in rats; whereas, in another long-term study in rats, wollastonite caused no tumors when injected into the peritoneal cavity. Thus, based on limited evidence of carcinogenicity in animals and inadequate human data, OTS is proposing to classify wollastonite as a possible human carcinogen (Category C). No other supporting evidence (e.g., genotoxicity data) of oncogenicity is available.

Available data are inadequate to evaluate the fibrogenic potential of wollastonite. A preliminary report of an NTP bioassay indicates no evidence of pulmonary fibrosis in rats

following chronic inhalation of wollastonite but data are not yet available for a full evaluation. Available epidemiological studies indicate a possible association between wollastonite exposure and some nonmalignant diseases such as impaired ventilatory capacity, mild fibrosis of the lung, pleural thickening and chronic bronchitis. However, because of a number of limitations, they do not provide convincing evidence of a causal relationship of nonmalignant respiratory diseases and wollastonite exposure. Nevertheless, these epidemiological findings do raise a health concern, particularly in view of positive results from *in vitro* cytotoxicity assays which are thought to be indicative of fibrogenic activity.

Overall, there is some evidence supporting a concern for a possible health hazard from exposure to wollastonite. However, it would appear that wollastonite is probably less hazardous than asbestos since available experimental data indicate that wollastonite is much less biologically active than asbestos. In order to fully assess the health effects of wollastonite, it is necessary to seek additional epidemiological studies and to fully evaluate the results of an inhalation bioassay recently completed by the National Toxicology Program.

6. Attapulgite

There is inadequate evidence of carcinogenicity of short-fibered attapulgite from available studies in humans. The results of a single small cohort study in the U.S. showed an excess of lung cancer among some groups of attapulgite workers. However, due to several limitations, this study did not provide

convincing evidence of a fiber etiology. Several experimental studies showed that short attapulgite fibers (<2 μm) in commercial use from the U.S., France, and Spain did not produce mesothelioma in rats by the intrapleural or intraperitoneal route. In addition, short attapulgite fibers from Spain did not induce tumors in rats following prolonged inhalation exposure. There is also no evidence of carcinogenicity in mice following life-time feeding with short-fibered attapulgite. These data are supported by negative findings from a single genotoxicity study on short attapulgite fibers. Short-fibered attapulgite is, therefore, not classifiable as to human carcinogenicity (Category D) on the basis of inadequate evidence of carcinogenicity from epidemiological and animal data.

In contrast, attapulgite samples from other geographical locations (e.g. Spain, U.K.) which contain considerable numbers of long fibers (>5 μm) have been shown to be tumorigenic in rats, causing the induction of lung tumors and mesotheliomas by inhalation, as well as pleural mesothelioma following intrapleural injection and abdominal tumors via the intraperitoneal route. Therefore, based on sufficient evidence of carcinogenicity in animals but in the absence of human data, OTS is proposing to classify long-fibered attapulgite as a probable human carcinogen (Category B2).

Available data have not provided evidence of fibrogenic effects for short-fibered attapulgite. The results of three studies in humans provide inadequate evidence of the development of nonmalignant respiratory diseases associated with exposure to short-fibered American attapulgite. The results of a long-term

animal study showed that short attapulgite fibers from Spain did not induce lung fibrosis in rats via inhalation. Morever, none of the available injection studies with short-fibered attapulgite from various geographical locations have reported any fibrotic lesions in treated rats. However, positive findings of several *in vitro* cytotoxicity studies suggest a possible fibrogenic concern for short-fibered attapulgite. In contrast, based on the positive results of a chronic inhalation study in rats with long-fibered attapulgite, it is concluded that attapulgite samples containing long fibers (>5 μm long) may induce lung fibrosis in humans.

In view of available findings, it would appear that the toxicological properties of attapulgite may depend on fiber length. Overall, there is insufficient evidence to support a health concern for short-fibered attapulgite in commercial use in the U.S. However, because these fibers are highly respirable, and appear to be biologically active in *in vitro*, adverse health effects remain a possibility. On the other hand, there is a reasonable basis to support a health concern for long-fibered attapulgite. Available animal data are not sufficient to allow a definitive assessment on the relative pathogenicity of long-fibered attapulgite compared to asbestos. However, since these fibers are not widely available for commercial use, they are not expected to pose significant health risks to humans. In order to fully assess the health effects of short-fibered American attapulgite, it is necessary to obtain additional epidemiological data and to conduct long-term inhalation studies in animals.

7. Aramid Fibers

There is no information available on the health effects of para-aramid fibers in humans. In the female rat, long-term inhalation of ultrafine para-aramid (Kevlar®) fibrils caused a dose-related production of lung tumors. Although there are no oncogenicity data on ultrafine para-aramid in animals via the intracavitary route, weak tumorigenic responses were observed in rats in two intraperitoneal injection studies with Kevlar® fiber and pulp containing a considerable number of fine fibrils. Thus, based on the sufficient evidence of the carcinogenicity in animals but in the absence of human data, OTS is proposing to classify ultrafine para-aramid as a probable human carcinogen (Category B2). There is no genotoxicity information available on ultrafine para-aramid.

Data from the same chronic inhalation study also indicate that ultrafine para-aramid (Kevlar®) is weakly fibrogenic in rats. The positive findings from an *in vitro* cytotoxicity study on short, thin Kevlar® fibers further support the concern for the fibrogenic potential of ultrafine para-aramid.

In view of these findings, it may be concluded that ultrafine para-aramid is potentially pathogenic. This fibrous material, however, does not pose a health risk to humans because it is not available in commerce. Available data, however, are not sufficient to provide definitive assessment on the comparative pathogenicity of ultrafine para-aramid to asbestos.

The positive results of two intraperitoneal injection studies in rats indicate that para-aramid pulps or fibers may have a low carcinogenic and fibrogenic potential. Thus, based on

the limited evidence of carcinogenicity in animals and in the absence of human data, OTS proposes to classify commercial grade para-aramid as a possible human carcinogen (Category C). Because of the generally nonrespirable characteristic of commercial grade para-aramid fiber and pulp, it would appear that the hazard potential of para-aramid is probably much lower than that of asbestos. However, it should be pointed out that since small numbers of para-aramid fibrils can result from peeling off the para-aramid fiber matrix and may become airborne, a possible health hazard may exist for exposure to para-aramid, particularly to the pulp form. In order to further assess the potential health effects of para-aramid, additional animal testing is recommended.

There are insufficient data to assess the health effects of Nomex® aramid fibers. Nomex® is not classifiable as to human carcinogenicity because of lack of data in humans and animals (Category D). Based on the fact that no effects were observed in a single long-term intratracheal instillation study in the rat, and that Nomex® is nonrespirable, it would appear that Nomex® poses no significant health hazard to humans. Because of a low health concern, no additional animal testing is recommended for Nomex®.

8. Carbon Fibers

There is no information available on the potential development of respiratory neoplasms in humans from exposure to carbon fibers. Furthermore, no data are available on the oncogenicity of carbon fibers in animals by inhalation. However,

carbon fibers were not found to induce tumors in rats following intratracheal instillation, intraperitoneal injection, or intramuscular implantation. The only studies that reported positive results were those from a subcutaneous study in which an increased production of local sarcomas was found in rats, and from a dermal bioassay demonstrating that benzene extracts of pitch-based carbon fibers were weakly oncogenic in mice. However, because there was no information available on the characteristics of the test materials, particularly particle size and morphology, the significance of these findings is questionable and the overall experimental evidence of carcinogenicity is considered to be inadequate. Carbon fibers are, therefore, not classifiable as to human carcinogenicity (Category D) on the basis of inadequate evidence from animal studies and in the absence of human data. The oncogenic potential of carbon fibers, however, is supported by available genotoxicity data which indicate that benzene extracts of pitch-based carbon fibers are clastogenic and induce DNA damage and repair. On the other hand, the evidence of clastogenicity of benzene extracts of polyacrylonitrile (PAN)-based carbon fibers is only suggestive.

There is inadequate evidence of fibrogenicity for carbon fibers. A small cross-sectional study conducted to date showed no evidence of pathological effects in the lungs of workers in a PAN-based carbon fiber production plant. With regard to experimental studies, there is no information available on the long-term inhalation toxicity of carbon fibers in animals. With the exception of one study which reported in an abstract that polyacrylonitrile (PAN)-based carbon fibers induced lung fibrosis

in rats via intratracheal instillation, several other animal studies showed that carbon fibers did not induce fibrosis in laboratory animals following subchronic inhalation exposure, intratracheal instillation, or intraperitoneal injection. Most of these studies, however, are of little value for the evaluation of the fibrogenic potential of carbon fibers because of limited scope, lack of particle size and morphology data of the test materials, and/or lack of details available on study design and findings. Furthermore, both negative and positive findings have been reported regarding the in vitro cytotoxicity of carbon fibers.

Although currently available data are insufficient to evaluate the potential health effects of carbon fibers, the data taken together suggest that carbon fibers do not appear to present a serious health hazard. Nevertheless, the marginally positive tumorigenic effects in a dermal study and the positive clastogenic effects in genotoxicity tests induced by pitch-based carbon fibers, suggest that a weak oncogenic potential for certain types of carbon fibers may exist. Because carbon fibers are much less respirable and less biologically active than asbestos, it would appear that they pose a lower degree of health hazard compared to asbestos. In order to further assess the health hazard of carbon fibers, it is necessary to seek results of an inhalation study now conducted at a private laboratory. Since the endpoint of this study is fibrosis, it is further recommended that a chronic animal study capable of detecting oncogenic effects be conducted if carbon fibers of respirable size enter the marketplace.

9. Polyolefin Fibers

There are no available epidemiological studies which examine the potential oncogenic effect of polyolefin fibers. Furthermore, there are no data available on the oncogenicity of polyolefin fibers in animals by inhalation. The results of an intratracheal insufflation study showed that both polyethylene and polypropylene fibers did not induce tumors in rats. However, the lack of information on the characteristics of the fibers, the dosages, and the specific methods of administration precludes any definitive assessment of the oncogenicity of these fibers under the conditions of the study. In a long-term intraperitoneal injection study in rats, polypropylene fibers were found to be weakly oncogenic. These results were only preliminary and a full evaluation cannot be made at this time. Therefore, polyolefin fibers are not classifiable as to human carcinogenicity (Category D) on the basis of inadequate evidence of carcinogenicity in animals and no human data.

No epidemiological studies have been conducted to determine the nonmalignant respiratory effects in humans from exposure to polyolefin fibers. There is no information available on the long-term inhalation toxicity of polyolefin fibers in animals. Available animal injection studies have provided inconclusive results. Polyethylene and polypropylene fibers did not induce fibrosis in rats in a long-term intratracheal insufflation study and in a short-term intraperitoneal injection study. These results are supported by the finding from a single *in vitro* study that polyethylene and polypropylene dusts exhibited very low cytotoxicity. However, the lack of information on the

characteristics of the test materials makes it difficult to draw any definitive conclusion on the fibrogenic potential of this fiber category. On the other hand, preliminary results of a long-term intraperitoneal injection study in rats with thin, long polypropylene fibers showed a strong degree of adhesions of the abdominal organs. However, in the absence of histological data, a full evaluation of this study cannot be made at this time. Overall, available data are inadequate to determine conclusively whether polyolefin fibers are fibrogenic. However, they seem to suggest a low fibrogenic potential for polyolefin microfibers.

In summary, available studies do not provide adequate data for a definitive assessment of potential health effects in humans exposed to polyolefin fibers by inhalation. However, the inhalation of polyolefin fibers or pulp may pose little or no health hazard because they are generally not respirable and would not be expected to produce lung diseases even if the material has some intrinsic activity. On the other hand, a possible health hazard potential may exist for polyolefin microfibers since they may be respirable. Additional animal testing is therefore recommended for polyolefin microfibers. Because of a low concern for the potential health effects of polyolefin fibers and pulps, further animal testing is not recommended at the present time.

I. Introduction

Human exposure to airborne asbestos fibers including amosite, chrysotile, and crocidolite has been associated with the development of malignant (e.g. lung cancer, mesothelioma) and nonmalignant (e.g. interstitial pulmonary fibrosis, also known as asbestosis) diseases. These diseases have also been induced experimentally in laboratory animals exposed to asbestos. As a result, concern has risen with the increasing development and use of other respirable fibrous substances. Nonasbestos fibers have come under considerable investigation primarily because they possess some asbestos-like characteristics (e.g. fiberlike morphology, dimensional range, durability) suspected to be important factors in the initiating of diseases. The objective of this report is to assess the human health effects associated with exposure to nonasbestos fibers and to evaluate the hypothesis that nonasbestos fibers may induce asbestos-like diseases.

The fibers under review comprise three categories: man-made mineral fibers (fibrous glass, mineral wool, ceramic fibers), naturally occurring fibers (erionite, attapulgite, wollastonite), and synthetic fibers (aramid fibers, carbon fibers, polyolefin fibers). These fibers were selected because of one or more of the following reasons: 1) they are commercially important; 2) they are considered potential asbestos substitutes; 3) they represent fiber types with broadly different physical and chemical characteristics; and 4) some health data are available on them.

This document reviews available data on pulmonary deposition, clearance and retention, in vivo toxicity, and in vitro biological activity of each of the nine fibrous materials. It also assesses the human health effects, primarily the potential development of malignant and nonmalignant respiratory diseases associated with inhalation exposure to each fiber, based on the combined available epidemiological and experimental evidence. Finally, it determines the adequacy of data for each of these fibers and makes testing recommendations to fill data gaps. A detailed review of the key epidemiological studies on the health effects posed by most of these fibers is presented in a separate document by Battelle (1988). Summaries and conclusions regarding human data have been derived from this report and are used in the overall hazard assessment of each fiber. The assessment of the carcinogenicity of fibers in humans is based on the U.S. EPA classification system for categorizing overall weight-of-evidence for carcinogenicity from human, animal, and supporting data (USEPA, 1986).

The last section of the document discusses overall findings about the whole fiber category and briefly evaluates the role of physicochemical properties of fibers in relation to biological activity and pathogenicity.

II. Man-Made Mineral Fibers (MMMF)

MMMF comprise three groups: fibrous glass, mineral wool, and ceramic fibers. MMMF have glassy structures rather than crystalline. Their length and diameter distribution differ

considerably and are dependent on the method of production and the chemical composition. In general, commercially produced MMMF are much coarser than asbestos fibers, although specialized samples have been produced with dimensions similar to those of asbestos. MMMF are usually coated with binding materials to produce fabricated shapes and forms. MMMF are monofilamentous, and thus do not split longitudinally into thinner fibrils, but may break transversely into shorter segments (NRC, 1984). Based on available data, the health effects of MMMF appear to vary substantially.

II.1. Fibrous Glass

Fibrous glass is made by forcing molten glass through an orifice, followed by air, steam, or flame attenuation. There are three major classes of fibrous glass: wool, textile, and special-purpose fibers. Glass wool fibers comprise approximately 90 percent of the total fibrous glass production and their major use is in thermal and acoustical insulation. They are typically 3-10 μm diameter but may range from 1-25 μm diameter, and therefore, may generate respirable airborne fibers. Textile fibers or continuous glass filament which account for 5-10 percent of the total fibrous glass are used in the manufacture of textile products and as reinforcements in plastics, rubber and paper. Textile fibers are, in general, nonrespirable because they have fairly large diameters with nominal diameters ranging from 6-15 μm. Special-purpose fibers with small diameters, representing less than 1 percent of fibrous glass production, are

manufactured for certain highly specialized uses in thermal insulation in aerospace vehicles and filter materials. This group includes fine fibers which have nominal diameters of 1-3 μm and very fine glass fibers (or microfibers) with diameters less than 1 μm. These fibers are highly respirable (NRC, 1984).

II.1.1. Fiber Deposition, Clearance and Retention

Available information regarding the inhalation, deposition, and clearance of glass fibers is fairly limited. The results of available studies suggest that fiber dimension is the most important factor in the deposition and elimination of glass fibers. Coarse glass fibers thicker than 1.5 μm are likely to be deposited mainly in the upper respiratory tract (nasopharyngeal and tracheobronchial regions) and would have little chance for alveolar deposition. Further, longer fibers (>10 μm) are less able to penetrate the alveolar region of the lung. Like other fibrous particles, glass fibers are probably eliminated rapidly from the upper airway via mucociliary clearance whereas fibers deposited in the alveolar space appear to be cleared more slowly, primarily by phagocytosis and to a lesser extent via translocation and possibly by dissolution. Short fibers (<5 μm) are believed to be removed mainly by macrophage uptake whereas longer fibers may be cleared at a slower rate by dissolution. In general, short fibers are cleared more rapidly than longer fibers, suggesting that fiber per fiber, short fibers are less likely to pose a toxicological concern.

The regional deposition of inhaled glass fibers has been studied by Morgan et al. (1980) and Morgan and Holmes (1984a). In these studies, rats were exposed for several hours by inhalation (nose-only) to glass fibers of different diameters (1.5 um or 3 μm) and lengths (5, 10, 30 or 60 um). The results of these studies showed that for fibers with 1.5 μm diameter and longer than 10 μm, fiber deposition in the lower respiratory tract and alveolar region was low and decreased with increasing fiber length. Moreover, alveolar deposition of thicker fibers (3 μm) was about one third of that of fibers of 1.5 μm diameter of similar lengths. These results, together with the previously reported data on other asbestiform mineral fibers (Morgan, 1979), indicated that alveolar deposition of fibers in the rat was optimal with an aerodynamics diameter of 2 μm, which is equivalent to an actual fiber diameter of approximately 0.5 μm. Available data also demonstrated that in general, increasing fiber length decreases the proportion of inhaled fibers deposited in the alveolar region (Harris and Timbrell, 1977; Harris and Fraser, 1976).

Immediately following deposition, there is a rapid decline in the lung content of glass fibers. Griffis et al. (1981) reported that 41-48 percent of lung burden of glass fibers in rats was cleared between daily exposures. The initial decline presumably represents early clearance from the upper respiratory airways, with a half time of less than one day. Fibers deposited in the upper airways are cleared by mucociliary activity which transports the fibers toward the oralpharynx. Fibers are then

swallowed, passed into the gastrointestinal tract and excreted into the feces. It has been shown that dogs excreted approximately 77 percent of the initial total burden of glass fibers within 4 days after inhalation exposure (Griffis et al., 1983). Similarily, in the rat, more than 95 percent of the total burden of glass fibers was associated with the gastrointestinal tract following a 2-hour exposure (nose-only), which was all excreted in the feces two days later (Morgan et al., 1980).

The elimination of fibers from the alveolar region is much slower than those in the upper airways via mucociliary clearance. The half time alveolar clearance of "TEL" glass fibers in the rat was reported to be approximately 44 days (Friedberg and Ullmer, 1984). Short fibers appeared to be cleared more efficiently than longer fibers. Morgan et al. (1982) showed that in the rat, more than 80 percent of glass fibers less than 5 um in length were cleared by one year following intratracheal instillation whereas no significant clearance of fibers greater than 10 um length could be detected over the same period. Bellmann et al. (1986) also found that short glass fibers ($\leq$5 μm) cleared faster than longer fibers (>5 μm) from the rat lung following intratracheal dosing. This study, however, showed that long glass fibers do clear from the lung while long crocidolite asbestos fibers (>5 μm) apparently do not clear from the rat lung over one year. On the other hand, long chrysotile asbestos fibers appear to split into fibrils as reflected by the observed increase in the number of fibers over a 6-month period.

Fibers are cleared from the alveolar region by a variety of mechanisms. The major pathway involves the removal of fibers by macrophage uptake. It is believed that fiber-laden macrophages (dust cells) move to the terminal bronchioles and are transported by the mucociliary system to the upper respiratory tract. These dust cells could then be swallowed. It would appear that the difference in the lung clearance between short and long fibers could be due to the fact that short fibers of less than 5 μm are efficiently removed by phagocytosis whereas the macrophage-mediated clearance is ineffective for fibers longer than 10 μm, due to the inability of macrophages to completely engulf the longer fibers (Bernstein et al., 1980; 1984; Morgan et al., 1982; Morgan and Holmes, 1984a).

The second pathway of fiber clearance from the alveoli involves the lymphatic system. Fibrous particles in the alveolar space are removed, either by macrophages or by themselves via an unknown mechanism, to the lymph nodes. The fate of the fibers in the lymph nodes is not known although they may escape the lymph nodes and enter the lymphatic and blood circulation, and may migrate to other tissues. There are few data available regarding the translocation of glass fibers. Glass fibers were found in the tracheobronchial and mediastinal lymph nodes of animals at different time periods after exposure to the mineral dusts by inhalation or intratracheal instillation (Lee et al., 1981; Bernstein et al., 1980, 1984; Wright and Kuschner, 1977). Furthermore, it appears that short fibers are more readily transported to the lymph nodes than longer fibers. In the study

by Morgan et al. (1982), measurements of the fiber content of the hilar lymph nodes of rats killed after one year following intratracheal instillation showed that approximately 4 percent of 5 um glass fibers had been transferred from the lung to the lymph nodes. Smaller proportions of the 10 um and 30 um fibers had been transported and no 60 um fibers were detected. With regard to the translocation of fibers to other organ tissues, only minimal amounts of glass fibers were found in the liver, spleen, and blood of animals exposed to the fibrous dust by inhalation (Lee et al., 1979, Griffis et al., 1983). Glass fibers were also detected in the spleen of rats after 2 years following intratracheal instillation. Further, Monchaux et al. (1982) reported recovery of fibers from all organs (blood, liver, kidney, brain) at 90 days after intrapleural injection of glass microfibers. However, increased pressure caused by this method of administration may have been partly responsible for these results.

It has been suggested that fibrous particles may also be cleared by dissolution. For glass fibers, the suggested evidence comes from morphological observations showing limited breakage and etching of the fibers retained over a long period following dosing, and chemical analysis of the recoverd fibers showing some changes of elemental composition (Johnson et al., 1984a; Le Bouffant et al., 1984; Spurny et al., 1983). These processes would result in shorter, thinner fragments which then could be cleared more efficiently by phagocytosis. The solubility of glass fibers in lung tissues appears to be dependent on fiber

size. In studies with rats, longer glass fibers dissolved more rapidly than shorter ones (Morgan et al., 1982; Morgan and Holmes 1984a; Bernstein et al., 1980, 1984). It has been suggested that the dependency of dissolution on fiber length may be due to differences in the intracellular and extracellular pH. The shorter fibers within macrophages are exposed to a lower pH of 7.17, while those outside are exposed to a higher extracellular pH of 7.4 (Morgan and Holmes, 1984a).

The solubility of glass fibers in lung tissues and in physiological fluids has been shown to be greater than that of amphibole fibers but may be similar or less then chrysotile (Forster, 1984; Spurny, 1983a; Spurny et al., 1983). The results of other in vitro studies also indicate that glass fibers have marked solubility rates in physiological fluids (Griffis et al, 1981; Leineweber, 1984; Klingholz and Steinkopf, 1984). Glass fibers of fine diameters degraded more rapidly than coarser ones (Spurny et al., 1983; Forster, 1984). Futhermore, the dissolution of long glass fibers (50 μm) in saline was much faster than that of short fibers (5 μm). These results indicate that the in vitro dissolution rate of glass fibers is proportional to the surface area of the fibers (Leineweber, 1984).

II.1.2 Effects on Experimental Animals

Fibrous glass has been extensively tested in laboratory animals for the ability to induce lung tumor, mesothelioma, and fibrosis. Information on the design and results of available

animal studies on glass fibers is summarized in Table 1 (pages 215-229). Such investigations have been conducted by several routes of exposure including inhalation, intratracheal instillation, intrapleural injection/implantation, and intraperitoneal injection. Animal exposure by inhalation represents the most relevant method for the assessment of risks to man. However, because of the technical difficulties and high costs, fewer long-term inhalation studies have been conducted in comparison with studies using the injection or implantation method which are more sensitive and generally more reproducible. Injection studies are of value in screening the test fiber for carcinogenicity and providing useful information regarding the intrinsic biological activity and carcinogenicity of the test fiber.

II.1.2.1 Oncogenicity

None of the available long-term studies have provided evidence of pulmonary or mesothelial carcinogenicity in animals exposed to fine glass fibers, glass microfibers or larger diameter glass fibers (e.g., glass wool) by inhalation. In contrast, many studies involving intrapleural or intraperitoneal administration of these fibers to animals have resulted in increases in mesothelioma of the pleura or peritoneum, respectively. In addition, two of several intratracheal instillation studies on glass microfibers also reported tumor induction with both lung tumors and mesothelioma in hamsters and lung tumors alone in the rat. By using the intrapleural implantation

method, Stanton and coworkers demonstrated that glass fibers less than 0.25 μm diameter and greater than 8 μm length have carcinogenic potential equal to that of asbestos fibers of similar size distribution. Other investigators also found that long, thin glass fibers are highly carcinogenic by the injection routes of exposure but are less effective than asbestos.

II.1.2.1.1 Inhalation Studies

The earliest studies with fibrous glass were those by Schepers and coworkers (Schepers and Delahunt, 1955; Schepers, 1955; Schepers, 1959a; Schepers, 1959b; Schepers, 1961) which were summarized in a final report in 1976 (Schepers, 1976). In one series of experiments, guinea pigs and rats were exposed to fairly large diameter glass wool fibers (average diameter close to 5 μm) at an average mass concentration of 0.145 mg/m^3 for 44 months, and at 0.03 mg/m^3 for 28 months, respectively. In another series of studies, guinea pigs, rabbits, rats, and monkeys were exposed to dust from two types of glass fiber reinforced plastics at either 3.8 mg/m^3 or 4.6 mg/m^3 for various time periods ranging from 8-24 months. No pulmonary tumors were reported in any exposed group. These studies, however, were inadequate to determine whether the fibrous products tested were carcinogenic in animals by inhalation due to 1) extremely low levels of fiber exposure, particularly for glass wool; 2) insufficient information on fiber size distribution in the dust cloud; and 3) poor survival of treated and unexposed animals.

Gross et al. (Gross et al., 1970; Gross, 1976) reported studies in which rats and hamsters were exposed for 2 years to a very high concentration of uncoated glass fibers (135 mg/m^3), glass fibers coated with a phenol-formaldehyde resin (106 mg/m^3), or glass fibers coated with a starch binder (113 mg/m^3). All three types of glass fibers in the dust cloud had an average diameter of 0.5 μm and an average length of about 10 μm. None of the rats or hamsters exposed to any of the fiberglass products developed lung or pleural tumors. However, it is not clear whether there was a sufficient number of animals at risk from late developing tumors due to a small number of animals and apparent poor survival of exposed animals. The survival pattern of unexposed control animals was not available for comparison.

Morrison et al. (1981) reported that 5 of 12 male A-strain mice developed bronchogenic or septal cell tumors 90 days after exposure to "crushed" glass insulation (80 percent were 6-11 μm long and 2-5 μm diameter) mixed in bedding material every 3 days for 30 days. However, the results of this study were inconclusive because of 1) insufficient information on the actual airborne glass fiber concentration; 2) lack of control animals caged in normal bedding; and 3) short exposure period and small number of exposed animals.

In 1979 and subsequently in 1981, Lee et al. reported studies in which rats and guinea pigs were exposed to glass fiber aerosol at an extremely high dust mass of 400 mg/m^3 for 90 days. The airborne dust particles had an average diameter of 1.2 μm and most particles were less than 2 μm long; thus, the

dust particles were predominantly nonfibrous. After 18 and 24 months post exposure, 2 of 19 rats and 2 of 8 guinea pigs developed bronchial alveolar adenoma while none of 13 rats and 6 guinea pigs exposed to clean air as controls had pulmonary tumors. Since these findings were based on a small number of animals, meaningful conclusions cannot be drawn from this study. Other limitations such as short exposure period further limit the conclusion that can be made about this study.

Goldstein et al. (1983, 1984) studied the effects of inhalation exposure of very fine fibrous glass in male baboons and compared them with the effects produced by crocidolite asbestos. Animals were exposed to a blend of Johns-Manville code 102 and code 104 glass microfibers (median diameter of airborne fiber of 0.6 um) at a mass concentration of 7.54 mg/m^3 (1,122 fibers/mL) for 35 months or UICC crodolite asbestos (median diameter of airborne fiber of 0.38 um) at a mass concentration of 15.8 mg/m^3 (1,128 fibers/mL) for 40 months. A total of 10 animals were used; the numbers in glass fibers exposed or positive control group were not specified. No neoplasms occurred with either of the dusts at 6-7 months following the end of exposure. Since the exposure and observation periods were short in relation to the lifespan of baboons, this study is considered inadequate for the evaluation of the oncogenic potential of fibrous glass. It should also be noted that neoplasms have been rare even in previous studies of asbestos-exposed monkeys and baboons.

Two studies by Wagner et al. (1984) and McConnell et al. (1984) were undertaken as a joint effort to compare the carcinogenic effects of glass microfibers with chrysotile asbestos, and to assess the comparability of results of similar inhalation studies at two different locations. In the study by Wagner et al. (1984), specific pathogen-free (SPF) male and female Fischer 344 (F344) rats (28 of each sex per group) were exposed for 3 or 12 months to coated or uncoated glass wool, glass microfibers, or UICC Canadian chrysotile asbestos. Chrysotile and glass microfibers were highly respirable with airborne fiber diameters ranging from 0.03 um to 2 um (mean diameter of 0.3 um) while glass wool had larger airborne fiber diameters ranging between 0.3 um and 3 um (mean diameter of 0.8 um). The respirable dust mass was 10 mg/m^3 in all cases. The concentrations of airborne respirable fibers (diameter <3 um, length >5 um) were 240 fibers/mL for uncoated glass wool, 323 fibers/mL for coated glass wool, 1436 fibers/mL for glass microfibers, and 3822 fibers/mL for chrysotile. Pulmonary response was assessed in rats sacrificed at 3, 12, and 24 months, and in animals that were allowed to live out their natural lifespan. One case of lung adenocarcinoma was found in animals exposed to glass wool with resin (1/48) and glass microfibers (1/48) while exposure to glass wool without resin resulted in one case of benign lung adenoma (1/48). In contrast, a total of 12 lung tumors (11 adenocarcinoma, 1 adenoma) were produced in the chrysotile group. All of the neoplasms were reported to occur

within 500-1,000 days after the start of exposure. Unexposed control animals developed no tumors.

Comparable results were obtained in the study by McConnell et al. (1984). In this study, male and female SPF F344 rats were exposed to the same glass microfibers or chrysotile asbestos preparation as used by Wagner et al. (1984), targeted at a respirable dust mass concentration of 10 mg/m^3 for 1 year. However, the actual cumulative dose of glass microfibers was approximately one half of that in the study by Wagner et al. (1984). Increased incidences of lung neoplasms were observed in 11 of 56 animals exposed to chrysotile but no tumors were found in the glass microfiber group (0/55). Two of 53 unexposed animals had lung adenocarcinoma. Most of the tumors were found after 24 months.

Analysis of the findings from these two inhalation studies showed that there was no statistically significant difference in tumor incidence between the unexposed controls and rats exposed to glass microfibers (Rossiter, 1982). In the study by Wagner et al. (1984), there was also no significant difference in tumor incidence between animals exposed to coated or uncoated glass wool fibers and the negative controls. However, both studies are limited with regard to study design including the use of a relatively small number of animals and short duration of exposure. Despite these limitations, these two studies demonstrated that while glass microfibers and glass wool fibers were not carcinogenic in rats under the conditions tested, similar mass concentration of chrysotile asbestos produced a

significant increase in the incidence of benign and malignant pulmonary neoplasms in the rat.

Smith et al. (1984, 1986) also found that glass microfibers and large diameter glass fibers caused neither lung tumor nor mesothelioma when inhaled by rats and hamsters. As a part of a comprehensive study, groups of male Syrian hamsters and female Osborne-Mendel rats (50-70 animals/group) were exposed "nose-only" for 24 months to one of the following dusts: (1) highly respirable glass microfibers (fiber product with mean diameter 0.45 um) at a mean mass concentration of 3.0 $\pm$ 0.6 mg/m^3 (approximately 3,000 fibers/mL) or 0.3 $\pm$ 0.1 mg/m^3 (300 fibers/mL); (2) fibrous glass "blowing wool" (fiber product with 3.1 um mean diameter) targeted at 10 mg/m^3 (100 fibers/mL); (3) flame attenuated fibrous glass (fiber product with 5.4 um mean diameter) at either 12 mg/m^3 (100 fibers/mL) or 1.32 mg/m^3 (10 fibers/mL); (4) fibrous glass insulation building (fiber product with 6.1 um mean diameter) at 9.0 mg/m^3 (25 fibers/mL). Positive control animals were exposed to UICC crocidolite asbestos at a mass concentration of 7 mg/m^3 (3,000 fibers/mL). One negative control group was exposed to clean air (sham controls). Following the exposure period, test and sham control animals were maintained for their natural lifespans. Another negative untreated control group remained in cages throughout their lives.

No primary lung tumors were found in rats or hamsters exposed to any of the fibrous glass dusts. On the other hand, one mesothelioma and two cases of bronchoalveolar tumors were detected in 57 asbestos-exposed rats. None of the hamsters exposed to

crocidolite asbestos developed lung tumors or mesothelioma. However, bronchoalveolar metaplasia, possibly a preneoplastic event in the development of epithelial tumors, was significantly elevated in hamsters exposed to crocidolite asbestos. With the exception of the occurrence of a bronchoalveolar tumor in a sham control hamster, none of the other sham control or unexposed control animals developed lung tumors. Thus, under the conditions of these lifetime studies there was no evidence of carcinogencity in rats or hamsters exposed to glass microfibers or large diameter fiberglass. The lack of significant tumorigenic response by crocidolite asbestos observed in this study might well be due to the use of a short-fibered material (approximately 95-97 percent were less than 5 μm long).

The long-term effects of inhalation of glass microfibers and glass wool were also studied in rats by Le Bouffant et al. (1984). Groups of 48 Wistar IOPS AF/Han rats (24 animals of each sex) were exposed to French commercial resin-free glass wool (Saint-Gobain), American produced glass microfibers (Johns-Manville code 100) or Canadian chrysotile asbestos at a respirable dust mass of approximately 5 mg/m^3 for 12 or 24 months. Because of differences in fiber size distribution and proportion of non-fibrous material present in the aerosols, the numerical concentrations of respirable fibers greater than 5 μm length as determined by optical microscopy were varied, ranging from 48 fibers/mL for glass wool (68 percent <1 μm diameter), 332 fibers/mL for glass microfibers (51 percent with diameters from 0.2-0.5 μm), to 5,901

fibers/mL for chrysotile (fiber diameter distribution not specified). The animals were sacrificed at 12, 24, and 28 months.

No pulmonary tumors were found in animals exposed to glass microfibers (0/48). A single lung tumor was observed at 24 months with the glass wool group (1/45). In contrast, nine cases of lung tumors were detected in the positive control group exposed to chrysotile (9/47). Negative control animals (unexposed) had no pulmonary tumors. Although there were no significant increases in lung tumors in rats exposed to either glass microfibers or glass wool fibers, this study is considered limited based on small numbers of animals and a relatively low level of fiber exposure. This study, however, demonstrated that under similar experimental conditions and mass concentrations, chrysotile asbestos was more potent in inducing lung tumors in rats than glass fibers.

Mitchell et al. (1986) also reported no evidence of pulmonary or mesothelial neoplasms in rats and monkeys following chronic inhalation of fibrous glass of varying geometry and mass concentrations. In this study, groups of F344 rats (50 animals of each sex per group) and male cynomolgus monkeys (15 per group) were exposed to (1) large diameter and long glass fibers with binder (4-6 um in diameter and >20 μm long) at approximately 15 mg/m^3 (Group I); small diameter and long glass fibers with binder (0.5-3.5 um in diameter and >10 μm long) at 15 mg/m^3 (Group II); (3) small diameter and long uncoated glass fibers (<3.5 μm in diameter and >10 μm long) at 5 mg/m^3 (Group III); and (4) small diameter and short uncoated glass fibers (<3.5 μm and <10 μm long) at 5 mg/m^3 (Group IV). Control animals (Group V) were exposed to

filtered air. The rats were exposed for a total of 86 weeks while the monkeys were dusted for only the 72 weeks. The animals were sacrificed following the termination of exposure.

Neither pulmonary tumors nor mesothelioma were detected in any treated monkey or rat groups. However, short treatment and study duration may have excluded observation of late developing tumorigenic effects, particularly in the monkeys. Furthermore, there was a low survival among treated and control rats. Approximately 37 percent of rats (187 of 500 animals) died spontaneously or were killed in a moribund condition before the termination of study. Many of the spontaneous early deaths were due to mononuclear cell leukemia (MCL). It was reported that there was a statistically increased incidence of MCL in each glass fiber exposed rat group. However, the investigators performed statistical analyses on the combined incidence in both males and females rather than analyzing the incidence data for the male and female populations separately. Reanalysis of data using Fisher exact test showed that for the male population only Group 3 ($p = 0.024$) and Group 4 ($p = 0.002$) displayed a significant increase in MCL. In the females, the incidence was significant only in Group I ($p = 0.047$). The biological significance of this finding remains uncertain since spontaneous increase in MCL is commonly seen in aged F344 rats.

II.1.2.1.2 Intrapleural Implantation/Injection Studies

Stanton and his colleagues reported a series of experiments (Stanton and Wrench, 1972; Stanton et al., 1977 and 1981) in which

they tested the ability of fibrous glass and other mineral fibers (including asbestos) of diverse dimensional distributions, to induce malignant neoplasms in female Osborne-Mendel rats by intrapleural implantation of the mineral dusts. Pledgets of coarse fibrous glass were coated with 40 mg of the test fibers suspended in gelatin and the pledgets were placed over the visceral pleura of the rats after open thoracotomy. Animals were observed for two years at which time survived animals were killed. The greatest increase in pleural sarcomas was observed for fibers with diameters less than 0.25 μm and lengths greater than 8 μm, although relatively high tumor yields were also produced with fibers having diameters up to 1.5 μm with lengths greater than 4 μm. These studies demonstrated that glass fiber with dimensional distribution similar to that of asbestos was equally carcinogenic as asbestos by intrapleural implantation.

Similar findings were obtained in the study by Smith et al. (1980) in which the tumorigenic effects of six fiberglass samples were tested in hamsters by intrapleural injection of a single dose of 25 mg of the test fiber. Intrathoracic tumors occurred in 9 of 60 animals which received fibers with a mean diameter of 0.1 μm and 82 percent longer than 20 μm. Fibers with a mean diameter of 0.33 μm and 46 percent longer than 20 um induced tumors in 2/60. Fibers with a mean diameter of 1.23 um and 34 percent longer than 20 μm also induced tumors in 2/60. No tumors were found in groups treated with the other three preparations containing fibers of similar diameter range but shorter lengths with only 0-2 percent longer than 10 μm. These results suggest that carcinogenicity is

associated with length and diameter of fibers; the thinner and longer the fiber, the more tumorigenic it is.

Wagner et al. (1973, 1976, and 1984) also tested the carcinogenicity of fiberglass of various types and size distributions by intrapleural injection in rats and confirmed that thin fiberglass was carcinogenic. Glass microfibers (Johns-Manville code 100), when injected as a single dose of 20 mg into the pleura of rats produced a significant increase in pleural tumors. In the 1976 study, 4 of 32 Wistar rats ($p = 0.01$) developed pleural mesothelioma while none of 32 control animals had tumors. Similarly, in the 1984 study, 4 of 48 Sprague-Dawley rats treated with glass microfibers developed mesothelioma. In contrast, coarse glass fibers (Johns-Manville code 110) produced no tumors in rats (Wagner et al., 1973, 1976) and only one case of mesothelioma was found among 48 rats injected with glass wool (Wagner et al., 1984). When comparing these results with those obtained with various types of asbestos in earlier experiments reported in Wagner et al. (1973) using identical intrapleural injection technique, finer glass fibers were considerably less carcinogenic than some of the asbestos samples, while coarse glass fibers were not tumorgenic.

Monchaux and coworkers (Lafuma et al., 1980; Monchaux et al., 1981) also reported induction of pleural tumors in rats following intrapleural injection of 20 mg of fine glass fibers (JM 104). The mean length of the test glass fibers was 5.8 μm and mean diameter of 0.229 μm. Pleural mesotheliomas were observed in 6 of 44 (14 percent) animals. Higher tumor incidences were produced by

UICC crocidolite (54 percent) and chrysotile (45 percent) asbestos while control animals receiving saline alone had no tumors. These findings were consistent with results of other intrapleural studies which showed that at a similar mass dose, crocidolite and chrysotile asbestos were more potent in inducing mesothelioma in rats than fine fibrous glass.

II.1.2.1.3 Intraperitoneal Injection Studies

In a series of studies, Pott and coworkers investigated the ability of fibrous glass to induce abdominal tumors in the rat by the intraperitoneal (i.p.) route of exposure. In the first series of experiments as reported in 1972, 1974 and 1976 (Pott and Friedrichs, 1972; Pott et al. 1974, 1976), a dose-related tumor induction (2.5 - 57.5 percent) was produced in female Wistar rats (40 per group) following intraperitoneal injection of a single dose of 2 or 10 mg, or 4 doses of 25 mg of fibrous glass (S + S 106; mean diameter 0.5 μm; 72 percent <5 μm long). Positive control animals receiving UICC chrysotile asbestos also developed tumors in the peritoneal cavity in a dose-related manner (15-67 percent). Histologically, nearly all the tumors from fibrous glass or chrysotile treated animals were sarcomatous mesothelioma. In both treated groups, the latency period for tumor development was inversely related to the dose of fibers injected. No tumors were observed in negative control animals receiving saline.

Similar results were obtained with uncoated glass fibers of type MN 104 (50 percent <0.2 μm in diameter; 50 percent <11 μm

long). This study showed a dose-related increase in mainly peritoneal mesothelioma in Wistar rats (80 animals/group) following intraperitoneal injection of the test fiber at a single dose of 2 or 10 mg, or 2 doses of 25 mg. Glass fibers of type MN 112 (50 percent <1 μm in diameter; 50 percent <28 μm long) produced a tumor incidence of 27.5 percent following an i.p. dose of 20 mg (Pott et al. 1976).

In a subsequent study, Pott et al. (1980) reported that intraperitoneal injection of 10 mg of glass microfibers (JM 104) to rats of 4 different strains resulted in different tumor rates ranging from 51 to 79.6 percent. The rat strains used in this study included Wistar (Ivanovas), SIV (Ivanovas), Sprague-Dawley (Hagemann), and Wistar (Hagemann). No other details of the experiments were available.

These results were confirmed in a later study by Pott et al. (1984) which showed a production of high incidences (40-70 percent) of abdominal tumors, primarily sarcoma or mesothelioma, in Wistar or Sprague-Dawley rats following intraperitoneal dosing with 2.5 or 10 mg of long glass microfibers (JM 104). Shorter glass microfibers (JM 100) induced lower incidences of tumors (2-10 percent).

Comparable findings were reported by other investigators using similar injection techniques. Davis (1976) injected into the peritoneal cavity of Balb/C mice and rats (strain unspecified) very fine glass fibers with an average diameter of 0.05 um as a single dose of 10 and 25 mg, respectively. Three of 25 mice and 3 of 18 rats developed peritoneal tumors. It was reported that

these tumors appeared identical to those produced in the peritoneal cavities of rats and mice by injection of crocidolite asbestos, as reported in earlier studies (Davis, 1974).

Recently, Smith et al. (1986) reported a 32 percent incidence of abdominal mesothelioma (8/25) in female Osborne-Mendel rats following an intraperitoneal injection of 25 mg of 0.45 um mean diameter fiber. UICC crocidolite asbestos produced tumors in 80 percent of the animals while no tumors were observed in saline controls or untreated animals.

II.1.2.1.4. Intratracheal Instillation Studies

Variable results on the carcinogenicity of fibrous glass via the intratracheal route have been reported. Tumor induction by fibrous glass was reported in one study by Mohr et al. (1984). In this study, groups of 136 male Syrian golden hamsters received eight weekly intratracheal instillations of 1 mg of the dusts. Thin fibrous glass (JM 104) of two different size lengths (mean diameter of 0.3 μm, and mean length of either 7 μm or 4.2 μm) and UICC crocidolite asbestos, were tested. Neoplasms, including lung carcinomas (4 percent), mesothelioma (27 percent) and thoracic sarcoma (4 percent), were found in hamsters treated with glass fiber samples at comparable rates. Interestingly, the incidence of mesothelioma in asbestos-treated animals was considerably lower than that of fiberglass. Control animals treated with titanium dioxide nonfibrous dusts developed no mesothelioma or lung carcinomas.

A subsequent intratracheal instillation study by the same group of investigators (Pott et al., 1987a) reported a low incidence of lung tumors in the rat treated with glass microfibers. Female Wistar rats were administered 20 weekly doses of 0.05 mg of JM 104/Tempstran 475 glass fibers (50% <3.2 μm long; 50% <0.18 μm in diameter). Five cases of lung tumors (1 adenoma, 2 adenocarcinomas, 2 squamous cell carcinomas) were found among 34 treated animals. In rats treated similarly with crocidolite asbestos (50% <2.1 μm long; 50% <0.20 μm in diameter), there were 11 cases of lung tumors out of 35 animals examined.

In contrast, several other intratracheal studies with fine glass fibers have not produced positive results. Gross et al. (1976) found no tumors in rats or hamsters injected intratracheally with multiple doses of uncoated glass fibers, glass fibers coated with resin, or starch binder. Glass fibers tested in this study had an average diameter of 0.5 μm and average length of 10 μm. Wright and Kuschner (1976 and 1977) also reported no tumor induction in guinea pigs injected with 12.5 mg of either thin, long glass fibers (90 percent >10 μm in length) or 25 mg of shorter fibers (90 percent <10 μm) of similar diameter (mean diameter <1 μm).

Recently, Feron et al. (1985) reported no mesothelioma or other tumors of the respiratory tract in Syrian golden hamsters treated with JM 104 glass microfibers (31 percent <0.25 μm in diameter; 89 percent <12 μm long) via intratracheal instillation (1 mg every 2 weeks for 52 weeks). Smith et al. (1986) also found no tumors in female Osborne-Mendel rats following

intratracheal instillation of 0.45 μm mean diameter glass fibers (1 mg weekly for 5 weeks).

II.1.2.2 Fibrogenicity

Fine fibrous glass and glass wool have been shown in several animal studies to produce minimal interstitial dust cell reaction without fibrosis following chronic inhalation. The pulmonary responses generally consist of macrophage infiltration with alveolar dust cell collections, alveolar proteinosis, and granuloma formation. In one study, fine fiberglass was reported to produce focal fibrosis in baboons. However, the small number of animals and the lack of unexposed animal control group limit the conclusions which can be made from this study.

In contrast, extensive pulmonary fibrosis has been induced in animals by intratracheal instillation and intrapleural injection of fine fibrous glass. Furthermore, marked peritoneal fibrosis has been produced via injection of fine glass fibers into the abdominal cavity of animals. The results of these injection studies showed that long, thin glass fibers are more fibrogenic than short, thin glass fibers, while thick glass fibers are apparently relatively inert, producing no significant pulmonary response. Pulmonary pathology induced by glass fibers by these routes of exposure, including inhalation, are much less severe than that produced by asbestos fibers in concurrent experiments.

Since the experimental details of most available studies are already presented in the discussion of oncogenicity and are

summarized in Table 1 (pages 215-229) only relevant information and test results on the fibrogenic effects are discussed in the following sections.

II.1.2.2.1 Inhalation Studies

Gross et al. (1976) found no development of pulmonary fibrosis in rats and hamsters exposed to very high dose levels (100 mg/m^3) of uncoated or coated glass fibers (mean length of 10 μm; mean diameter of 0.5 μm). The survival rate of treated animals, however, was poor. Schepers et al. (1976) also reported that glass wool and glass fiber reinforced plastics did not induce fibrosis in rats, guinea pigs, rabbits, and monkeys, following a two-year inhalation exposure to various concentrations of the dusts (0.03-4.6 mg/m^3). This study also had a high incidence of mortality.

In 1979 and subsequently in 1981, Lee et al. reported that the major pathological lesion found in rats, hamsters, and guinea pigs which were exposed for 90 days to a very high glass fiber dust cloud (400 mg/m^3) with a full lifespan follow up, was alveolar proteinosis. Very slight alveolar interstitial fibrosis occurred in a few old animals. It should be noted that these experiments used fibers of small aspect ratios (3:1) with only 7 percent of the fibers considered fibrous in shape. Furthermore, the exposure period was relatively short.

Morissett et al. (1979) reported that a group of 20 male albino mice which were exposed to respirable glass fiber (<3 μm diameter and <10 μm length) at 1,070 fibers/mL for six weeks did not develop pulmonary fibrosis. This study is considered limited because of the short duration of exposure.

In 1980, Johnson and Wagner reported that electron microscopic examination of lung tissues of rats (two SPF Fischer rats/group) exposed to 10 mg/m^3 of glass microfibers, resin coated glass wool or uncoated glass wool for 50 weeks revealed focal fibrosis. However, pulmonary fibrosis was not found in the more extensive investigation by Wagner et al. (1984). Fischer 344 rats developed minimal interstitial cellular reaction (grade 3.4) following a one-year exposure to glass microfibers (10 mg/m^3) and a one-year follow up. Animals exposed to glass wool either with or without resin at similar mass concentration (10 mg/m^3) had no significant pulmonary responses (grade 2.6 and 2.4, respectively). On the other hand, animals exposed to UICC Canadian chrysotile showed evidence of early interstitial fibrosis (grade 4.1).

McConnell et al. (1984) also found no evidence of pulmonary fibrosis in Fischer 344 rats following a one-year exposure to very fine JM 100 glass fibers, obtained from the same source as described in Wagner et al. (1984). Animals which were exposed to UICC crocidolite developed mild pulmonary fibrosis. It should be pointed out the accumulative exposure of glass fibers in this study was only half of that achieved in Wagner's study.

Le Bouffant et al. (1984) found only minimal pulmonary response in rats after one year exposure to fine fibrous glass or glass wool at 5 mg/m^3. They were limited to alveolar and interstitial macrophage reactions, with mild septal fibrosis in the case of glass microfibers. In the case of chrysotile fibers, only hyperplastic changes of the alveolar lining were observed; pulmonary fibrosis was not detected.

Smith et al. (1986) exposed groups of rats and hamsters to fine fibrous glass (0.45 um mean diameter; 300 or 3,000 fibers/mL) and coarse fibrous glass (>3 μm diameter; 25-150 fibers/mL) for 2 years. Pulmonary fibrosis was not found in any of the treated animals (50-60 animals/group) with fibrous glass. Many of the rats and hamsters exposed to UICC crocidolite asbestos, however, developed prominant pulmonary fibrosis.

Baboons were reported to develop focal peribronchiolar fibrosis following exposure to respirable glass microfiber dust clouds for 35 months at 1,122 fibers/ml (7.54 mg/m^3). The test fibers had a mean diameter of 0.5 um and median length of 6 um. Pulmonary lesions induced by glass microfibers were morphologically similar to those produced by crocidolite asbestos; however, the incidence and severity in unexposed control animals were not reported (Goldstein et al., 1983).

In the study by Mitchell et al. (1986), cynomolgus monkeys and Fischer 344 rats were exposed via inhalation to dust clouds of fibrous glass of varying geometry and concentrations (5 or 15 mg/m^3) for 18 months and 21 months, respectively. There was no evidence of lung fibrosis in either species. Both species had

pulmonary macrophage aggregates and granulomas containing fibrous glass. The rats had grossly visible pleural plaques which were not seen in the monkeys. No positive controls were included in this study.

II.1.2.2.2 Intratracheal Instillation Studies

Kuschner and Wright (1977) reported that long, thin glass fibers (92 percent >10 μm in length; <1 μm in diameter) produced a marked fibrotic reaction in the guinea pigs following intratracheal injection of a single dose of 12 mg. The instillation of short, thin glass fibers (93 percent <10 μm) at similar doses produced only macrophage aggregation in the alveoli.

In the study by Pickrell et al. (1983), groups of 20 male Syrian hamsters were intratracheally instilled with one of two uncoated glass microfibers (2 and 7 mg; 0.1-0.2 μm diameter) or one of three commercial glass fiber samples (17-21 mg; 2.3-4.1 μm diameter), or UICC crocidolite (6 mg; 0.24 μm diameter). The thinner glass microfiber (0.1 um) caused deaths from pulmonary edema shortly after instillation. Increased collagen deposition and mild pulmonary fibrosis were observed in animals treated with the thicker glass microfiber (0.2 μm) and one type of commercial glass fiber (2.3 m) at 11 months after instillation. However, the microfibers produced a greater effect than the commercial type, while crocidolite asbestos induced the greatest response. No pulmonary responses were observed in animals treated with the

other two types of commercial glass fibers which had larger diameter (3-4 μm).

Marked lung fibrosis was also found in 27 percent of female Osborne-Mendel rats (7/22) treated intratracheally with 2 mg of 0.45 μm mean diameter glass fibers once a week for five weeks compared to saline control animals. However, the incidence of pulmonary fibrosis in positive control rats instilled with crocidolite asbestos was much higher and the lesions were more severe (Smith et al., 1986).

II.1.2.2.3. Intrapleural Injection Studies

The relationship between fiber dimension and fibrogenicity was also demonstrated by Davis (1976). Groups of 25 Balb/c mice received 10 mg of one of 4 samples of glass fibers of varying lengths and diameter, by the intrapleural route. Short fiber samples (<20 μm) of both large (3.5 μm) and small diameter (0.05 μm) produced only small discrete granulomas with minimal fibrosis. Long fiber samples (>100 μm) produced massive fibrosis, which was comparable to that induced by asbestos.

II.1.2.2.4 Intraperitoneal Injection Studies

Pott et al. (1974) reported that glass fibers (average diameter of 0.5 μm; 72 percent less than 5 μm in length), when injected into the abdominal cavity of rats as a single dose of 50 mg, produced marked peritoneal fibrosis. The effect was less extensive at lower doses (2 and 10 mg). Similarly, Smith et al. (1986) found extensive peritoneal fibrosis in female Osborne-

Mendel rats injected intraperitoneally with 25 mg of 0.45 μm mean diameter glass fibers.

II.1.3. In Vitro studies

II.1.3.1 Genotoxicity

Several studies on glass fibers have been performed, ranging from bacterial mutation tests to transformation studies in mammalian cells. Most studies dealt with Code 100 and Code 110 fiberglasses of various lengths. These two glass fibers differ in their diameters with mean diameters of roughly 0.12 μm and 1.9 μm, respectively. Glass fibers do not appear to induce gene mutations in bacterial cells, although this evidence is very limited. The two major effects that consistently appear with Code 100 fiberglass are aberrations and transformation in cultured cells. The cytogenetic effects seen by Code 100 fiberglass appear to be less effective than chrysotile asbestos, although sometimes comparable to crocidolite asbestos. Other cytogenetic effects, such as induction of sister chromatid exchanges (SCE) and micronuclei, were not seen; again however, only a very limited number of studies were available. Code 110 fiberglass does not appear to have effects comparable to Code 100 fiberglass.

II.1.3.1.1. Mutational Effects

Glass fibers and several asbestos samples were tested in two bacterial mutation tests (Chamberlain and Tarmy, 1977). The two glass fibers examined were Code 100 (mean length of 2.7 μm, mean

diameter of 0.12 μm) and Code 110 (mean length of 26 μm, mean diameter of 1.9 μm) fiberglasses. The asbestos samples included UICC Canadian chrysotile, UICC crocidolite as well as a "cleaned crocidolite" (Magnetite), UICC amosite, UICC anthophyllite and SFA chrysotile. All samples were found negative in the Salmonella/mammalian activation test in strains TA1535 and TA1538 with and without activation. The asbestos samples were tested at 0.1 and 1.0 mg/plate, but the glass fiber concentrations were not specifically stated. Similar negative results were found in the E. coli mutation test with strains B/r, WP2, WP2 uvrA and WP2 uvrA pol A. All samples were tested up to 1,000 μg/plate in the E. coli test, except for Code 110 fiberglass (100 μg/plate top dose). The authors suggest that the negative results may be due to a lack of phagocytosis of fibers by bacteria (bacteria apparently do not phagocytize). Also, in general, bacteria appear resistant to the cytotoxic effects of fibers whereas mammalian cells are sensitive. Despite the problems of not specifically stating the concentrations for glass fibers and the lack of data using other strains (e.g. TA98, TA100) in the Salmonella assay, it appears that bacterial systems may not be appropriate to assay the potential mutagenicity of fibers.

II.1.3.1.2. Chromosomal Effects

Glass fibers were examined in several studies for chromosomal effects and were compared to the effect induced by chrysotile and/or crocidolite asbestos. The first study to examine the potential effects of glass fiber and glass powder in

Chinese hamster ovary (CHO) cells was reported by Sincock and Seabright (1975). They found that chrysotile and crocidolite at 0.01 mg/mL both induced polyploid cells, cells with fragments, and other chromosomal changes (such as breaks and double minutes) as well as an increase in the percentage of abnormal cells. Glass fiber and glass powder at 0.01 mg/mL appeared to cause no effect different from controls. However, an effect by specifically sized glass fibers cannot be ruled out by this study, as the dimensions of the glass fibers were not specified.

In subsequent studies, Sincock (1977) and Sincock et al. (1982) examined the potential chromosomal effects of glass fibers in CHO cells in more detail. Asbestos samples, including chrysotile, crocidolite, amosite, and anthophyllite were also examined. Chrysotile and crocidolite up to 0.1 mg/mL for 48 or 72-hour cell exposure consistently induced high levels of chromosome damage including increases in breaks, dicentrics, inversions, rings, percent abnormal cells, and polyploidy. Code 100 (up to 0.1 mg/mL; lengths 2.7-26 μm) induced a significant increase in the same parameters, but at levels usually less than that for chrysotile and crocidolite. It produced polyploidy at levels similar to amosite and anthophyllite. Code 110 (lengths 2.7-26 μm), glass powder (coarse borosilicate) and glass of 2 μm diameters but of specific lengths (<10 μm, 25 μm, 50 μm and 100 μm) all had no effect.

Sincock et al. (1982) also examined the potential effect of the asbestos and glass fiber samples (described above) on cultured human cells. Five different fibroblast cell strains

(not exceeding passage 15) and two different lymphoblastoid cell lines were used. No increase in chromosomal damage was noted for any sample assayed. The authors also searched for fiber-induced micronuclei in the lymphoblastoid lines, but noted no increase over controls. It was shown in this report that cultured CHO cells sustained damage to fibers, but the cultured human cells were not overtly damaged. There is no apparent explanation, although the authors suggest that the difference may be due to differences in excision repair, the nature of the transformed lines, or the species of origin. The phenomenom seems similar for the asbestos and glass fibers tested by these authors.

Code 110 fiberglass and its respirable fraction were tested in Chinese hamster lung (V79) cells (Brown et al., 1979b). The fibers were milled to lengths <200 μm. The respirable fraction was obtained from collecting respirable dust and was designated 110R. Code 110 had 9×10^9 fibers/g and 110R had 25.2×10^9 fibers/g of which a 20 μg/mL concentration for both was tested in the V79 cells. The 110R sample presumably had similar diameters as 110, but had shorter lengths. In comparison to a crocidolite positive control, 110 fibers had no observed chromosomal effect. 110R however, induced increased fragments, breaks, and percent of cells with abnormal spreads over the negative control. The 110 sample was weakly cytotoxic to V79 cells, but the 110R sample induced noticeable cytotoxicity.

Oshimura et al. (1984) examined the cytogenetic effects of several asbestos and fiberglass samples on tertiary cultures of Syrian hamster embryos. Chrysotile induced time- and

concentration-dependent increases in frequencies of cells with numerical changes (aneuploidy and tetraploidy), chromosomal aberrations (breaks, fragmentation, exchanges, dicentrics), binuclei, and micronuclei after 24- and 48-hour treatments (concentrations at 2 $\mu g/\mu m^2$). Code 100 fiberglass and crocidolite had similar effects (48 hour exposure; 2 $\mu g/\mu m^2$), but both of these were less effective in inducing cytogenetic changes than chrysotile. Code 110 fiberglass and alpha-quartz (non-fibrous mineral dust) were without significant effects. Milling of code 100 samples reduces the length (not given) as well as the cytogenetic effect. This suggests that the appropriate length, not the chemical composition, of Code 100 fiberglass is responsible for the cytogenetic effects seen in this study.

Glass fiber and asbestos samples were tested for their ability to induce sister chromatid exchanges (SCE) in CHO and cultured human cells (Casey, 1983). Chrysotile, crocidolite, Code 100, and Code 110 fiberglasses were added to cell cultures under two regimes. Concentrations of 0.001, 0.01 and 0.05 mg/mL were added to cells one hour after seeding in procedure one, and cells in suspension were exposed to 0.01 mg/mL before seeding in procedure two. By either treatment, no fiber tested induced SCE over the control frequency in CHO cells, human lymphoblastoid cells, or primary human fibroblasts (8-10 passages). No cell cycle effect was seen in the fibroblasts under either procedure and a mitotic delay was seen in CHO cells only with procedure two (except for Code 110). It appears that while glass fibers may

induce aberrational and ploidy type changes in cells, glass fibers do not induce SCE, at least based on this study.

II.1.3.1.3. Transformation Effects

Two studies report results from transformation studies using asbestos and glass fibers. In the earlier report, Sincock (1977) described the effect of chrysotile, crocidolite, and coarse fiberglass (Code 110) on murine 3T3 cells. The two asbestos fibers (0.01 mg/mL) induced foci indicative of transformation after only 7 days of exposure. Coarse fiberglass had no apparent effect.

Hesterberg and Barrett (1984) examined the effect of many more fiber samples on tertiary cultures of Syrian hamster embryos. Chrysotile and crocidolite produced linear increases in transformation frequency (concentrations 0.25-2 $\mu g/cm^2$) with chrysotile more potent than and twice as cytotoxic as crocidolite. Extracted chrysotile actually induced a 3-fold higher frequency than unextracted samples, indicating that possible contaminating organics may not have a role in fiber-induced transformation. Code 100 fiberglass (2 $\mu g/cm^2$; 9.5-16 μm length) was as active as chrysotile in transforming cells. Milled Code 100 (0.95-1.7 μm length) and milled chrysotile both exhibited greatly reduced transformation and cytotoxic activities. The reduction is presumably due to the reduction in fiber length. Code 110 fiberglass was less toxic than Code 100 and was 20-fold less potent than Code 100 for transformation. Two nonfibrous mineral dusts (alpha-quartz and Min-U-Sil) also

induced concentration dependent increases in transformation, but at much higher concentrations (10-75 $\mu g/cm^2$) than chrysotile and Code 100 fiberglass. They were also much less toxic than chrysotile. The authors suggest that the slopes of the response curves indicate a one-hit mechanism for transformation that would suggest a direct effect by the fibers.

II.1.3.2. Cytotoxicity

Fibrous glass was tested in several *in vitro* studies to determine its cytotoxic potential in various cell culture systems. Glass microfibers were found highly cytotoxic to lung and peritoneal macrophages, P388D1 macrophage-like cells, phagocytic ascites tumor cells, Chinese hamster lung fibroblast V79-4 cells, rabbit lung fibroblasts, type II human alevolar A549 cells, and human bronchial epithelial cells. The cytotoxic effect of thin glass fibers appears to approach that of asbestos fibers. On the other hand, coarse fiberglass (code 110) had little or no cytotoxic effects although its respirable fraction was found to have some cytotoxic activities in one study. These studies also indicated that long, thin glass fibers are more cytotoxic than short, thin fibers. This may be due to the fact that long fibers are incompletely phagocytized and, as a result, may damage the cell membrane and may cause subsequent release of enzymes, followed by cell death. The ability of fibrous glass to cause cellular membrane damage, as measured by hemolysis of red blood cells, has been reported, but a varying degree of hemolytic activity was found.

II.1.3.2.1. Erythrocytes

Available data on the hemolytic activity of glass fibers are limited. Jaurand and Bignon (1979) reported that glass fibers had a poor hemolytic effect, compared to UICC chrysotile, when incubated with human red blood cells. However, the hemolytic activity of glass fibers was similar to that of UICC crocidolite. In contrast, Ottolenghi et al. (1983) showed that Pyrex glass fibers (dimensions not specified) at 100 μg/mL did not cause hemolysis in chicken erythrocytes. Amosite also had no effect, while chrysotile asbestos induced high hemolytic effect. In an abstract, Nadeau et al. (1983) reported that glass microfibers (diameter of 0.2 μm; length of 221 μm) induced marked hemolytic activity in rat erythrocytes. No further details were available for a conclusive evaluation.

II.1.3.2.2. Phagocytic cells

Tilkes and Beck (1983a) examined the cytotoxic effects of glass microfibers of different size distributions in guinea pig and rat lung macrophages. The release of lactate dehydrogenase (to demonstrate plasma membrane permeability) and beta-glucuronidase (to indicate lysosomal permeability) were measured as indicators of cytotoxicity. It was shown that macrophage toxicity (100 μg/mL) was length dependent; the highest toxicity was seen with fibers longer than 5 um. For fibers of similar physical size dimension, the cytotoxic effects of amosite and crocidolite asbestos and glass fibers were equivalent when doses were gravimetrically equivalent. Glass microfibers (JM 100) also

caused a significant depression in phagocytic activity of macrophages. Phagocytosis was assayed quantitatively by determining the amount of luminescence produced after the addition of serum-opsonized zymosan A particles.

These results confirmed the earlier findings in the study by Beck et al. (1972) showing that fine fiberglass (0.25-1.0 μm diameter; 1-20 μm length) induced an increase in cell membrane permeability of guinea pig alveolar and peritoneal macrophages, as measured by increased release of lactate dehydrogenase and lactate levels.

Pickrell et al. (1983) tested the in vitro cytotoxicity of two uncoated glass microfiber insulation materials (0.1-0.2 μm diameter), three types of fibrous glass-containing household insulation (2-4 μm diameter) and crocidolite asbestos (0.25 um diameter) in pulmonary alveolar macrophages isolated from Beagle dogs. It was reported that the most cytotoxic of the fibers tested was crocidolite asbestos. Household insulations were not cytotoxic at the highest concentration tested. Both types of glass microfibers had cytotoxicities intermediate between household insulation and crocidolite asbestos.

In an abstract, Nadeau et al. (1983) also reported that long glass microfibers (0.2 um x 221 μm) were highly cytotoxic to rat pulmonary alveolar macrophages. However, the experimental details were not provided in this report.

Brown et al. (1979a) studied the cytotoxicity of JM code 100 glass microfibers (nominal diameter of 0.05-0.09 μm) and JM code 110 glass fibers (nominal diameter of 1.5-2.49 μm) in mouse

peritoneal macrophages. Code 100 microfibers were more cytotoxic than code 110 fibers as reflected by 2-3-fold differences in the release of lactate dehydrogenase and beta-glucuronidase levels at 160 μg/mL. Similar findings were obtained by Davies (1980) who demonstrated that fine glass fibers (JM 100) at a concentration of 160 μg/mL were cytotoxic toward mouse peritoneal macrophages. Coarse glass fibers code JM 110 had no cytotoxic effects but the respirable fraction of JM 110 glass fibers had some cytotoxic activities.

Tilkes and Beck (1980, 1983b) investigated the cytotoxicity of fibrous glass in phagocytic ascites tumor cells derived from Wistar rats, as measured by the release of lactic dehydrogenase and the inhibition of cell proliferation as determined by cell count, DNA, RNA, and protein synthesis. It was found that for long glass fibers (>20 μm), the thinner the fiber, the greater the toxicity in this cell culture system. In addition, a glass fiber fraction with comparable geometry to a UICC chrysotile asbestos fraction exhibited the same high cytotoxicity.

In the study by Lipkin (1980), borosilicate glass fiber (dimension unspecified) which was obtained from the same lots used by Stanton et al. (1977) was found highly toxic to P388D1 macrophage-like cells at a concentration of 100 μg/mL. The cytotoxicity of glass fibers in this cell culture was well correlated with potency in pleural sarcoma induction reported by Stanton et al. (1977) in intrapleural implantation studies.

II.1.3.2.3. Nonphagocytic cells

Brown et al. (1979b) studied the effect of glass wool on Chinese hamster lung fibroblasts (V79-4 cells) and human alveolar type II (A549) cells. Respirable fractions of coated or uncoated glass wool produced a dose-dependent inhibition of cell growth (10-50 μg/mL) of V79-4 cells. Uncoated glass wool also produced a significant increase in the number of giant cells when added to the A549 cell cultures at 200 ug/mL.

Chamberlain et al. (1980) reported that code 100 glass microfibers (dimensions and concentrations not provided) reduced the colony forming ability of V79-4 cells and induced giant cells in A549 cell cultures. In contrast, code 110 coarse glass fibers had no effect. The actual data were not provided in this study to fully evaluate the findings.

In a study by Richards and Jacoby (1976), glass fibers (dimensions unspecified) caused a slight cytotoxicity to rabbit lung fibroblasts when added to cell cultures at 50 μg/mL. Fiberglass also induced morphological changes and alterations in reticulin deposition in the fibroblast cultures. In contrast, UICC chrysotile asbestos was highly cytotoxic to fibroblasts and caused more extensive morphological changes in these cultures at a similar mass concentration.

Glass fibers (1-100 μg/mL) were also found to induce a dose-dependent inhibition of clonal growth rate of human bronchial epithelial cells (Haugen et al., 1982). In this cell culture system, UICC chrysotile was more cytotoxic than glass fibers by more than 100-fold.

II.1.4. Assessment of Health Effects

Existing studies have provided no clear evidence of a carcinogenic or fibrogenic hazard in humans. However, available animal studies show that fine fibrous glass is carcinogenic and fibrogenic by the injection route of exposure. Thus, there remains a concern for possible health hazards from inhalation exposure to fine fibrous glass, i.e., fibers with diameters less than 3 μm. A low health concern is also raised for exposure to glass wool, which does contain some respirable fine fibers. As for textile fibers (continuous glass filaments) which are generally nonrespirable, they would appear to pose little hazard to exposed humans. On the basis of available animal data, it is concluded that all fiberglass categories appears to be less pathogenic than asbestos.

II.1.4.1 Oncogenicity

Available health and toxicological information seems to indicate that the oncogenicity of fibrous glass varies for the three major categories i.e., fine fibrous glass, glass wool, and continuous glass filament. The variable oncogenic potential for these classes of fibrous glass appear to be related to their different fiber size distributions.

By using the U.S. EPA weight-of-evidence criteria for carcinogenicity (USEPA, 1986), fine fibrous glass and glass wool may be categorized as possible human carcinogens (Category C) on the basis of inadequate evidence of carcinogenicity in humans and limited evidence in animal studies. On the other hand, continuous glass filament is not classifiable as to human carcinogenicity (Category D) due to inadequate evidence of carcinogenicity from epidemiological and animal data.

Available data from recent cohort studies suggest that workers engaged in the manufacture of glass wool and small diameter fibers might be at increased risk of developing respiratory cancer. Small excesses of respiratory cancer death have been observed among workers exposed to glass wool and small diameter glass fibers but no excess of respiratory cancer has been found among glass filament workers. A dose-related trend has not been found although it should be noted that exposure to fibrous glass has been extremely low. The causal relationship between fibrous glass exposure and the development of respiratory cancer is therefore not considered credible at this time. There is also inadequate evidence of an increased mortality from mesothelioma in available MMMF cohorts. On the basis of available information, the weight of evidence of carcinogenicity of fibrous glass, i.e., glass wool, continuous glass filament, and small diameter fibers (fine fiberglass), from studies in humans is considered inadequate. Since the results of relevant epidemiological studies on fibrous glass have been reviewed in details and assessed in a report by Battelle (1988), only a brief description of the study design and findings are presented here.

Enterline et al. conducted a large cohort study on fibrous glass workers from 11 plants in the U.S.. These workers had at least one year exposure between 1945 and 1963. For those working in facilities where small diameter fibers were prevalent, the criterion was greater than six months of exposure. The cohort's mortality experience was traced through 1977 in the early study (Enterline et al, 1983) and in the subsequent studies mortality

was followed to 1982 (Enterline et al., 1986; 1987). The average level of exposure to glass fibers (<3 μm in diameter) for all fiberglass plants was 0.039 fibers/mL.

In this study, a slight excess of mortality from respiratory cancer was observed among glass wool workers which was nonsignificant based on local rates but was statistically significant compared to national rates. Mortality from respiratory cancer increased nonsignificantly with time from exposure but was not related to duration of exposure, cumulative exposure, or average intensity of exposure. In the glass filament subcohort, there was no excess of respiratory cancer and no upward trend with time since first exposure, duration of exposure, or average intensity of exposure. Among workers in 4 fiberglass plants ever exposed to small diameter glass fibers (<3 μm), there was a nonsignificant excess of respiratory cancer mortality which increased nonsignificantly with more than 30 years since onset of exposure. However, the small number of deaths limits any definitive conclusion regarding the relationship between fine fiberglass exposure and respiratory cancer. The results of a nested-case control study using respiratory cancer cases among fibrous glass (type unspecified) workers showed a statistically significant association between respiratory cancer and smoking but not between respiratory cancer and cumulative fiber dose (Enterline et al., 1986; 1987).

Similar results were obtained in the European study. Simonato et al. (1985; 1986a; 1986b) also performed a historic cohort investigation of glass wool workers from five plants and

continuous glass filament workers from two facilities in Europe. This study cohort consisted of men and women employed with at least one year of employment from 1933-46. Mortality was followed to 1982 and risks were also examined for early, middle and late production phases. In the glass wool cohort, there was no overall excess of lung cancer deaths by using the local mortality rates but there was a small nonsignificant excess when compared to national rates. Mortality from lung cancer increased nonsignificantly with time since first exposure but was not related to duration of exposure or to different technological phases, reflecting differences in the intensity and quality of exposure. Among glass filament workers there was no excess of lung cancer and no upward trend with time since first exposure or duration of exposure.

A third study by Shannon et al. (1986) reported a significantly elevated risk for lung cancer in a small Canadian glass wool cohort. However, analyses of lung cancer deaths by duration of employment and time since first exposure indicated no consistent dose-related trends.

There were no excessive mesothelioma deaths reported in the two large cohort studies on MMMF workers exposed to fiberglass and mineral wool. Simonato et al. (1985) observed one death due to mesothelioma in the European study of 24,000 workers. Enterline et al. (1986) reported two mesothelioma deaths in a cohort of 16,000 workers followed for 36 years. However, an investigation by Engholm et al. (1986) reported an excess number of mesothelioma in the Swedish construction industry. The study population

consisted of 135,000 male workers exposed to MMMF (no distinction between exposure to fibrous glass and mineral wool). There was a significantly increased mortality from pleural mesothelioma in the Swedish cohort. However, possible confounding by asbestos exposure and several limitations of the study (e.g. exposure defined by job category and no monitoring data to define categories) limit the conclusions that can be made about this finding.

Experimentally, there is insufficient evidence for the carcinogenesis of fibrous glass in animals by inhalation. Fine fibrous glass (including glass microfibers) and glass wool have been tested in several long-term inhalation studies, in several animal species including the rat (Wagner et al., 1984; McConnell et al., 1984; Le Bouffant et al., 1984; Smith et al., 1986), hamster (Smith et al., 1986), monkey (Mitchell et al., 1986) and baboon (Goldstein et al., 1983). There was no statistically significant increase in the incidence of lung tumors or pleural mesothelioma in any of these studies; only a few tumors of the respiratory tract occurred in some experiments in rats (Wagner et al., 1984; Le Bouffant et al., 1984). Although none of the available inhalation bioassays is considered adequately studied, collectively they do demonstrate that at equal mass concentrations and similar experimental conditions, chrysotile asbestos generally induced significant increases in lung tumors while fine fibrous glass and glass wool did not cause significant tumorigenic responses in laboratory animals following chronic inhalation exposure.

However, data from studies in which glass fibers were administered by nonphysiological routes indicate that a carcinogenic hazard potential does exist for glass wool and fine fibrous glass, in particular, for glass microfibers which contain a considerable number of long, thin fibers. Glass wool, fine fibrous glass and glass microfibers were not found to cause tumors in a number of intratracheal instillation studies in rats (Smith et al. 1986; Gross et al., 1976), hamsters (Feron et al., 1985; Gross et al., 1976) and guinea pigs (Wright and Kuschner, 1976, 1977). However, in one study, lung tumors and pleural mesotheliomas were observed in hamsters by intratracheal instillation of glass fibers with a median diameter of 0.3 μm in hamsters (Mohr et al., 1984). In another study by the same laboratory, lung tumors were also induced in rats instilled intratracheally with glass microfibers (Pott et. al., 1987a). In studies where various samples of glass microfibers (<1 μm diameter) were tested by intrapleural implantation (Stanton et al., 1977, 1981) or injection (Smith et al., 1980; Wagner et al., 1976, 1984; Monchaux et al., 1981) variable incidences of pleural tumors were induced in rats. Furthermore, peritoneal mesotheliomas or sarcomas were found in the abdominal cavity in rats following intraperitoneal injection of glass microfibers (Davis, 1976; Pott et al., 1974, 1976, 1980, 1984). By the intrepleural route, glass wool also caused low incidences of mesothelioma in a few studies (Stanton et al., 1981, 1977; Wagner et al., 1984) while other studies have produced no mesothelioma (Wagner et al., 1973; 1976). Stanton and coworkers also demonstrated that glass fibers less than 0.25 μm

diameter and greater than 8 μm length have carcinogenic potential equal to that of asbestos fibers. Similarly, other investigators found that long, thin glass fibers are highly carcinogenic by the injection routes of exposure but are generally less effective than asbestos at equal mass doses.

The relevance of the injection method with regard to human exposure is considered questionable considering that it bypasses normal physiological deposition and clearance mechanisms in the respiratory tract. Positive results from studies using intrapleural or intraperitoneal injection/implantation method in the absence of positive findings from inhalation experiments do not indicate that these fibers will produce tumors in man upon inhalation. However, positive results from such injection studies as found in the case of fine glass fibers and glass wool indicate that they have the potential to induce tumors when introduced to the target tissues in sufficient quantity. Furthermore, the fact that in two studies involving intratracheal instillation of small doses of glass microfibers (to mimic the inhalation exposure condition) resulted in the induction of tumors distal to the administration site (lung tumors and mesothelioma) indicate that fine fiberglass can reach the critical target tissues (lung and pleural mesothelium) if a sufficient amount of fibers can penetrate the upper respiratory airways. Whether or not these materials when inhaled will indeed reach the target tissues in sufficient quantity to cause tumors depends on the respirability characteristics of the fibers, which are not operative in the

injection study. Thus, in the absence of positive findings from available inhalation studies, the weight-of-evidence for the carcinogenicity of fine fibrous glass and glass wool in animal studies is considered limited.

In vivo animal data are supported by positive findings from a few genotoxicity studies showing that fine glass fibers appear to have similar genotoxic effects (clastogenicity and transformation) as asbestos (Sincock et al., 1982; Oshimura et al., 1984; Hesterberg and Barrett, 1984).

There are no studies available that examined the carcinogenicity of glass filaments in animals via inhalation. Moreover, large diameter glass fibers did not produce mesothelioma in rats via the intrapleural route (Stanton and Wrench, 1972; Stanton et al., 1977; 1981). Thus, the weight-of-evidence of carcinogenicity for glass filament in animal studies is considered inadequate.

II.1.4.2. Fibrogenicity

There does not appear to be any convincing evidence for increased risks of non-malignant respiratory disease (NMRD) from exopsure to fibrous glass. There is also no definitive evidence for the development of lung fibrosis in animals inhalation. However, the positive findings from several injection studies in animals and in vitro cytotoxicity studies indicate that fine fibrous glass may be fibrogenic.

Available epidemiological studies have provided weak or no evidence of excess mortality from NMRD in fibrous glass workers.

In the large cohort study in the U.S. (Enterline et al., 1986; 1987; Enterline, 1987), there was no significant excess mortality from NMRD among glass wool workers compared with local rates, although there was a statistically excess based on national rates. However, there was no relationship with time from first exposure, or duration of exposure. Among glass wool workers "ever exposed" to small diameter fibers, there was no excess of NMRD mortality but there was a slight nonsignificant increase with time since first exposure. In the continuous glass filament cohort, no excess NMRD mortality was observed based on either local or national rates.

Other investigators have not observed an association of NMRD and fibrous glass exposure. In the large European study (Simonato et al., 1985; 1986a; 1986b), there was no excess mortality from NMRD in the glass wool or continuous filament cohort, nor was there a relationship with time from first exposure or duration of exposure. Shannon et al. (1986) also did not find an excess risk of NMRD in their study of Canadian glass wool workers. A deficit in risk for NMRD was reported by Engholm et al. (1986) in their study of Swedish workers exposed to MMMF (fibrous glass and mineral wool).

The results of a respiratory morbidity study (Weil et al., 1983) showed some evidence of radiographic opacities in the lung of a limited number of fibrous glass workers. However, this study showed no evidence of pulmonary fibrosis, no increase in respiratory symptoms, and no impaired lung function.

Overall, there is inadequate experimental evidence of fibrogenicity for fine fiberglass and glass wool via inhalation exposure. It has been shown in several studies that chronic inhalation exposure to fine glass fibers or glass wool produced only minimal interstitial dust cell reaction without fibrosis in rats (Wagner et al., 1984; Smith et al., 1986; McConnell et al., 1984; Mitchell et al., 1986; Le Bouffant et al., 1984), hamsters (Smith et al. 1986) and monkeys (Mitchell et al., 1986). One study reported the development of focal fibrosis in baboons exposed to fine glass fibers (Goldstein et al., 1983). However, the small number of animals and the lack of unexposed control animals limit the conclusions that can be made from this study.

In contrast, more extensive pulmonary fibrosis was induced in animals by intratracheal instillation (Wright and Kuscher, 1977), Pickrell et al., 1983; Smith et al., 1986) or intrapleural injection (Davis, 1976). Furthermore, intraperitoneal injection of fine glass fibers generally resulted in marked peritoneal fibrosis (Pott et al., 1974; Smith et al., 1986). The results of these injection studies also indicate that long, thin glass fibers are more fibrogenic than short, thin fibers, while thick glass fibers (>3 μm diameter) are relatively inert. In general, glass fibers produced less severe and less progressive pulmonary lesions than those induced by asbestos via either inhalation or injection at equal mass concentrations or doses.

The *in vivo* findings are further supported by results from several *in vitro* studies showing that fibrous glass is cytotoxic to various cell types in culture. The cytotoxicity of fibrous

glass was found to be a function of fiber dimension, with longer (>10 μm), thinner (<1 μm) fibers being most cytotoxic, whereas coarse fibrous glass with fairly large diameter fibers (>3 μm) were less cytotoxic (Tilkes and Beck, 1980, 1983a, 1983b; Brown et al., 1979a; Davies, 1980; Pickrell et al., 1983).

II.1.5. Recommendations

Although glass wool and fine fibrous glass have been tested extensively, none of available inhalation studies are considered to be adequate. In addition, there are no data available regarding the comparative dose-response effects with asbestos. Thus, it would be useful to conduct additional long-term animal inhalation or injection studies on fibrous glass. Further epidemiological studies are also necessary to clarify the pathogenicity of fibrous glass in humans.

II.2. Mineral Wool

There are two major types of mineral wool, i.e., rock wool and slag wool. Rock wool is made by melting natural igneous rocks and then drawing, blowing, or centrifuging the melts into fibers. Slag wool is produced by a similar process from blast furnace slag. Mineral wools are primarily used for thermal insulation but are also used for sound dampening and reinforcement of other materials. Slag and rock wools have nominal diameters ranging from 6-9 μm, but also contain a relatively high proportion of respirable fibers (diameter $\leq$3 μm). Thus, mineral wools are likely to generate respirable airborne fibers during production and processing (ICF, 1986).

II.2.1. Fiber Deposition, Clearance and Retention

There is limited information on the pulmonary deposition, clearance, and retention of mineral wool fibers. However, the results of available animal studies suggest that mineral wool deposition and clearance are dependent on both fiber length and diameter. The lung clearance of mineral wools appears to be rapid soon after inhalation, presumably via the mucocillary system and by phagocytosis. Translocation of inhaled mineral wool fibers to regional lymph nodes and abdominal organs appears to be limited. During later periods, mineral wools are eliminated slowly, presumably by dissolution.

Hammad (1984) studied the pulmonary deposition and clearance of mineral wool in the rat. A group of 49 male rats were exposed "nose only" to aerosol of mineral wool (fiber type not specified)

at 300 fibers/mL for 6 consecutive days. Count median length and median diameter of airborne fibers were 13 μm and 1.2 μm, respectively. Rats in groups of seven were sacrificed at 5, 30, 90, 180, and 270 days after the last day of exposure. The pulmonary deposition of fibers, as approximated by fiber retention after 5 days of clearance, appeared to be dependent on both fiber length and diameter. Fibers of diameters less than 1.3 μm and shorter than 50 μm were found much more frequently in the lung than thicker and longer fibers. The pulmonary clearance of mineral wools was multiphasic. During the 5-30 and 30-90 day periods, mineral wool had a half-life of 38 days, whereas the half-lives increased to 68 days and 175 days for the 90-180 and 180-270 day periods, respectively. Approximately 3-7 percent of mineral wool fibers deposited in the lung were retained after 270 days. Most of the fibers which were retained in the lung had fairly large diameters (1.0-1.3 μm) and were relatively short (<5 μm).

Fiber retention has also been demonstrated in the rat lung following long term inhalation exposure to mineral wools (Johnson et al., 1984a; Le Bouffant et al., 1984). In the lung, rock wool fibers were found predominantly in the alveolar or interstitial macrophages (Johnson et al., 1984a). Fiber translocation from the lung to the tracheobronchial glands and diaphragm has been observed for rock wool as evidenced by the presence of small amounts of fibers in these organs (Le Bouffant et al., 1984). Migration of slag wool fibers outside the lung to the abdominal organs including the spleen, liver, and diaphragm have also been

reported after instillation of fibers into the trachea of rats, hamsters, and rabbits (Spurny et al., 1984).

Mineral wool fibers appear to dissolve in the rat lung after a long period of a time (1-2 years) following inhalation exposure (Hammad et al., 1985; Johnson et al., 1984a; Wagner et al., 1984), or via inoculation into the trachea (Morgan and Holmes, 1984b; Spurny et al., 1984). Apparent fiber dissolution in abdominal tissues has also been observed after injection of mineral wool fibers into the peritoneum of various rodent species (Pott et al., 1984; Spurny et al., 1984).

Slag and rock wools appear to have different solubility properties. Morgan and Holmes (1984b) showed that rock wool appeared to dissolve relatively slowly in the lung and that dissolution apparently occurred more rapidly at the end of fibers than in the middle, where the diameter was essentially unchanged. Similar findings have been obtained by other investigators reporting that etching of fibers in the lung was minimal for rock wool (Johnson et al., 1984a; Wagner et al, 1984). On the other hand, slag wool fibers in the lung (Spurny et al., 1984) and in abdominal tissues (Pott et al., 1984; Spurny et al., 1984) showed a considerable degree of corrosion. Analyses of the retained slag wool fibers by various spectroscopic methods revealed a partial to complete loss of alkali metals and alkaline earth ions (Spurny et al., 1984).

Results from *in vitro* experiments which studied the dissolution of mineral wools in simulated physiological fluid, are in agreement with the *in vivo* observations on fiber durability.

Slag wool fibers have been shown to undergo rapid and extensive leaching in physiological fluid, while the leaching of rock wool fibers was much less extensive (Forster, 1984; Klingholz and Steinkopf, 1984). Leineweber (1984) also demonstrated that in physiological saline, mineral wool fibers dissolved at a slow rate, but it is not clear whether the tested fibers in this study were rock wool or slag wool.

II.2.2. Effects on Experimental Animals

A number of studies have been conducted to evaluate the tumorigenic and fibrogenic effects of mineral wools in laboratory animals. The experimental protocols and findings of available studies are summarized in Table 2 (pages 230-233).

II.2.2.1. Oncogenicity

The results of available long-term studies have not provided evidence of pulmonary or mesothelial carcinogenicity in rats or hamsters exposed to mineral wool fibers by inhalation (Wagner et al., 1984; Le Bouffant et al., 1984; Smith et al., 1986). On the other hand, malignant mesothelioma of the pleura or peritoneum have been produced in rats following either intrapleural (Wagner et al., 1984) or intraperitoneal (Pott et al., 1984, 1987b) injection of various types of mineral wool at varying yields. However, significantly more neoplasms were found in animals exposed to asbestos fibers by either inhalation or injection route of exposure at equal mass concentrations or doses.

II.2.2.1.1. Inhalation studies

In the study by Wagner et al. (1984), a group of 56 SPF Fischer 344 male and female rats were exposed to a dust cloud of rock wool (58 percent <1 um diameter; 64 percent >10 μm long) at a mass concentration of 10 mg/m^3 (equivalent to approximately 227 fibers/mL) for 12 months. Of the 48 exposed animals which were allowed to live out their full lifespans, there were two cases of lung tumors (one benign adenoma and one adenoma with some malignant features). Unexposed control animals had no tumors. Taking into account the known occurrence of spontaneous species-specific lung neoplasms in F344 rats, there were no significant differences in tumor incidence between unexposed and exposed rats. In contrast, UICC chrysotile asbestos produced 12 cases of lung neoplasms (11 adenocarcinomas, 1 adenoma with some malignant features).

In a limited inhalation study by Le Bouffant et al. (1984), a group of male and female Wistar IOPS rats were exposed to rock wool at concentration of 5 mg/m^3 of dust for 24 months. The rock wool fibers had fairly large diameters (only 22.7 percent <1 μm). Because the rock wool fiber dust also contained a large proportion of nonfibrous particles and fragments that did not conform with the definition of a fiber, the numerical rock wool fiber concentration was very low (11 fibers/mL) in comparison with that of Canadian chrysotile (9,978 fibers/mL) used as control. No tumors were found in the exposed males (0/24) or female (0/23) rats. On the other hand, nine cases of lung tumors were observed

in the chrysotile exposed animals at 24 months. Unexposed control animals had no lung tumors.

In a study reported by Smith et al. (1986),-female Osborne-Mendel rats and male Syrian hamsters were exposed "nose-only" to a mineral wool dust cloud at a mass concentration of 12 mg/m^3 (200 fibers/mL) for 24 months. The tested mineral wool fibers had fairly large diameters with a median value of 2.7 um. No primary lung tumors were found in the exposed hamsters (0/69) and rats (0/55). Tumors were not observed in any of the untreated control rats (0/125) or hamsters (0/112) nor in rats exposed to clean air (0/59), and only one bronchoalveolar tumor (1/58) was found in the control hamsters group exposed to air. No significant production of neoplasms were found in positive control hamsters (0/58) or rats (3/57; 1 mesothelioma, 2 bronchoalveolar tumors) exposed to UICC crocidolite asbestos. It was believed that the lack of a significant tumorigenic response by crocidolite asbestos observed in this study could have been due to the use of relatively short-fibered material (95 percent <5 μm long).

II.2.2.1.2. Intrapleural Injection Studies

Wagner et al. (1984) injected 20 mg of either rock wool (with or without resin) or slag wool (with or without resin) into the pleural cavity of groups of 48 SPF Fischer 344 rats. Approximately 70-80 percent of rock wool and slag wool, either with or without resin, were less than 5 um long and less than 1 um diameter. Three cases of mesotheliomas were produced in the animal group treated with rock wool with resin, and two cases by

rock wool without resin. The slag wool produced no mesotheliomas while UICC chrysotile asbestos induced mesothelioma in six animals. It should be pointed out that the injected rock wool and slag wool dusts contained more nonfibrous particles (i.e., aspect ratio less than 3) than fibrous particles.

II.2.2.1.2. Intraperitoneal Injection Studies

Pott et al. (1984) reported very low tumor yields in female Sprague-Dawley and Wistar rats (40-60 animals per group) following intraperitoneal injection of mineral wools. A single dose of 5 mg of slag wool containing a high proportion of very thin fibers (90 percent <0.28 μm diameter) and long fibers (9 percent >10 μm length) produced only a tumor rate (sarcoma or mesothelioma) of 5 percent, which was not statistically different from zero. Rock wool of fairly large diameter fibers (50 percent <1.9 μm diameter) produced a 16 percent incidence of sarcoma/mesothelioma of the peritoneum of rats treated with three doses of 25 mg of dust, while no neoplasms were observed in rats after 15 months following a single dose of 10 mg of thin rock fibers (50 percent <0.64 μm in diameter). Basalt wool (50 percent <0.52 μm diameter, 50 percent <5.8 μm long) also did not produce neoplasms in female Wistar rats after 15 months following a single i.p. dose of 5 mg.

Recently, Pott et al. (1987b) reported that relatively thick basalt wool produced a high incidence of abdominal tumors in rats following repeated intraperitoneal injections. In this study, female Wistar rats received five weekly injections of 15 mg of basalt wool (50 percent <1.8 μm diameter; 50 percent <20 μm

long) suspended in saline. The animals were kept for their entire lifespan. Peritoneal mesothelioma/sarcoma were found in 30 of 53 treated animals. High tumor yields were also obtained with UICC/Canadian chrysotile asbestos at considerably lower doses, 11/36 at 0.05 mg, 21/34 at 0.25 mg, and 30/36 at 1.0 mg). The tumor incidence in negative saline controls was 1/102.

II.2.2.2. Fibrogenicity

The experimental data on the fibrogenic effect of mineral wools are limited. The majority of studies showed a lack of fibrogenic response following chronic inhalation exposure. However, the results of a limited inhalation study suggest that a low fibrogenic potential may exist for mineral wools.

In a limited study by Johnson and Wagner (1980), groups of two SPF Fischer 344 rats were exposed to dust clouds of either respirable rock wool (length >5 μm) or UICC chrysotile B asbestos, at 10 mg/m^3 for 50 weeks. The animals were sacrificed 4 months following the inhalation period. Rock wool and chrysotile asbestos produced focal fibrosis. However, the effect was more marked following inhalation of chrysotile than after exposure to rock wool. Two unexposed rats had normal lungs.

On the other hand, lung fibrosis was not observed in the more extensive study subsequently reported by Wagner et al. (1984). Rats which were exposed to rock wool dust clouds (10 mg/m^3) for 12 months developed interstitial cellular reactions to the dusts without fibrosis (grade 3) at 12 and 24 months upon sacrifice or following spontaneous death. In contrast, animals which were

exposed to UICC chrysotile asbestos showed evidence of early interstitial fibrosis (grade 4) similar to those seen in human asbestosis. Unexposed control animals had normal lungs (grade 1).

A lack of fibrogenic effects was also reported in the chronic inhalation study by Smith et al. (1986). Hamsters and rats did not develop lung fibrosis following a 2-year "nose-only" exposure to mineral wool fibers (12 mg/m^3). Similarly, there was no evidence of lung damage in rats exposed to Saint-Gobain rock wool for 24 months at 5 mg/m^3 as reported by Le Bouffant et al. (1984).

II.2.3. In Vitro Studies

II.2.3.1. Genotoxicity

There is no information available on the genotoxicity of mineral wools. Thus, the genotoxic potential of mineral wools cannot be assessed at the present time.

II.2.3.2. Cytotoxicity

The results from two *in vitro* studies indicate that rock wool and slag wool are cytotoxic to phagocytic and nonphagocytic cells. In general, mineral wools appear to be less cytotoxic than crocidolite asbestos.

Davies (1980) studied the cytotoxic effect of rock wool, slag wool, and UICC crocidolite asbestos on mouse peritoneal macrophages. The macrophages were exposed to 160 µg/mL of the respirable fraction of the test fibers (fiber dimension unspecified). Cytotoxicity was measured by determining the release of lysozomal enzyme beta-glucuronidase (BGD) and

cytoplasmic enzyme lactic dehydrogenase (LDH). Both slag wool and rock wool caused a significant release of BGD (2.5- and 1.8-fold, respectively) when added to the cell culture. Rock wool also induced a 1.7-fold increase in the release of LDH, while UICC crocidolite asbestos produced a 3.2- and 2.9-fold increase in LDH and BGD levels, respectively. It was also reported that removal of binder material from rock and slag wools had no effect on their activity; however, no data were presented to support this conclusion.

Brown et al. (1979b) studied the effects of respirable fractions of rock and slag wools (size distribution unspecified) on Chinese hamster (V79-4) lung cells and human alveolar epithelial type II lung tumor cells (A549). Both slag and rock wools, with or without resin (10-50 ug/mL) caused a dose-dependent cytotoxic response toward V79-4 cells, but the uncoated samples were slightly more cytotoxic than the equivalent resin coated samples. Uncoated slag and rock wools, when added to the A549 cell cultures at 200 ug/mL, produced a significant increase in the formation of giant cells, i.e., clusters of 200 cells or more. The effect of UICC crocidolite asbestos on A549 cells, however, was much greater than that of mineral wools.

Nadeau et al. (1983) reported in an abstract that mineral wools were more or less unreactive toward rat pulmonary alveolar macrophages and rat erythrocytes. However, no other information was provided to evaluate these findings.

II.2.4. Assessment of Health Effects

Data from available epidemiological and experimental studies indicate that mineral wools are potentially carcinogenic and possibly fibrogenic. Thus, there is a reasonable basis to conclude that mineral wool fibers may present a health hazard to exposed humans. However, based on available experimental data, mineral wools appear to be much less biologically active than crocidolite asbestos in a few cytotoxicity studies and less carcinogenic than chrysotile asbestos fibers in a few limited inhalation studies, and therefore would pose a health hazard of less magnitude than that of asbestos.

II.2.4.1. Oncogenicity

By using the weight-of-evidence criteria for carcinogenicity, mineral wool may be classified as a probable human carcinogen (Category B1) on the basis of limited evidence of carcinogenicity from epidemiological studies and limited evidence from animal studies.

The earlier epidemiological evidence relating mineral wool exposure as reviewed by the National Research Council (NRC, 1984) suggests an association with respiratory cancer. More recent data from two large cohort mortality studies (Enterline et al., 1986; 1987; Simonato et al., 1985; 1986a; 1986b) now indicate that mineral wool workers are at increased risk of developing respiratory cancer. Overall, there are no large excesses of deaths from respiratory cancer in any of available studies. Evidence supporting an etiological relationship between respira-

tory cancer and mineral wool exposure includes the consistently elevated respiratory cancer risks seen in several mineral wool plants from different countries, and the higher risks in workers who have had 20 or more years elapse since first exposure. In addition, in a nested-case control study in which confounding by smoking was controlled, there was a weak but significant trend observed between mineral wool exposure and respiratory cancer. However, consistent dose-response relationships have not been observed among available studies. It should be pointed out that the low levels of exposure in nearly all plants studied and the potential exposure misclassification could have contributed to the apparent lack of dose-response relationships. On the basis of available findings, the weight-of-evidence for a causal association between exposure to mineral wool and occurrence of respiratory cancer is considered limited (Battelle, 1988).

The results of key epidemiological studies on mineral wools have been reviewed in detail in a report by Battelle (1988). Briefly, the studies by Enterline et al. examined deaths due to respiratory cancer (malignant neoplasms of the bronchus, trachea and lung) among male mineral workers from 6 plants in the United States. These workers were employed for at least 1 year from 1941-63. In the early study (Enterline et al., 1983), the cohort was followed from 1941-1977. An update of the study included a 5 additional years of follow up from 1946 through 1982 (Enterline et al., 1986; 1987). The mean fiber exposure level in the mineral wool plants was 0.3 fibers/mL. It was found that respiratory cancer death was significantly elevated using both national and

local mortality rates. Analyses of data by duration of exposue, cumulative exposure and average intensity of exposure showed no dose-related trend. However, a significant excess was observed for 20 or more years from first exposure. The investigators also conducted a small case-referent study which controlled for cigarette smoking. It was found that there was a statistically significant relationship between fiber exposure and respiratory cancer for mineral wools even after smoking was considered (Enterline et al., 1987).

Simonato et al. (1985; 1986a; 1986b) conducted an historical cohort study on mineral wool workers from seven rock/slag wool facilities in 4 European countries. The study was followed from the time that production started (1937-1943) through 1982. The study cohort consisted of men and women with at least one year employment. There was a nonsignificant elevated increased risk of lung cancer mortality rates among rock/slag wool workers compared to national and regional rates. There was no relationship between mortality from lung cancer and duration of exposure. However, significant excess of lung cancer death was found after more than 20 years follow up among workers first exposed during the early technological phase, i.e., before the introduction of oil binders and presumably dustier conditions existed. Exposure such as smoking or other occupational substances are considered unlikely to provide a sufficient explanation for the excess of lung cancer risk. However, possible effects of such exposure either alone or in combination with fiber exposure cannot be excluded on the basis of available information.

The experimental evidence for the oncogenicity of mineral wool is considered to be limited. In two long-term inhalation studies in which rats were exposed to rock wool, no statistically significant increase in lung tumor incidence was observed (Wagner et al., 1984; Le Bouffant et al., 1984). In a third study in which rats and hamsters were exposed chronically to mineral wool (fiber type not specified) containing fairly large diameter fibers, no lung tumors were found (Smith et al., 1986). Although available studies have not provided evidence of oncogenicity via inhalation exposure, it should be noted that due to a number of experimental limitations, none of the studies on mineral wool are considered adequately studied. However, rock wool was shown to produce a low incidence of pleural mesothelioma in rats via intrapleural injection (Wagner et al., 1984) and both rock wool and basalt wool induced abdominal tumors following intraperitoneal injection (Pott et al., 1984, 1987b). Slag wool, on the other hand, produced no pleural tumors in rats by intrapleural inoculation (Wagner et al., 1984) and only a statistically nonsignificant increase in peritoneal tumors via intraperitoneal injection (Pott et al., 1984). In contrast, at equal mass concentrations or doses chrysotile asbestos induced high incidences of lung tumors in rats via inhalation (Le Bouffant et al., 1984; Wagner et al., 1984) and pleural mesothelioma by intrapleural injection (Wagner et al., 1984).

There were no reports available that examined the genotoxicity of mineral wools.

II.2.4.2. Fibrogenicity

The epidemiological evidence of an association between increased risks of nonmalignant respiratory diseases and mineral wool exposure is considered inadequate. However, since there is limited evidence from experimental studies indicating that mineral wool may have some cytotoxic and fibrogenic effects, there remains a concern for possible development of pulmonary fibrosis associated with mineral wool exposure.

No increased mortality from nonmalignant respiratory diseases was found for the European rock wool workers (Simonato et al., 1985, 1986a, 1986b). However, in the U.S. study (Enterline et al., 1986; 1987; Enterline, 1987), a statistically nonsignificant excess of deaths from nonmalignant respiratory diseases was observed among mineral wool workers based on local or national rates, but the data did not establish a relationship with interval from onset of employment nor was there a dose-related trend using exposure indices including duration of employment, cumulative level of exposure, and average intensity of exposure. Furthermore, the results of a morbidity study (Weill et al., 1983) showed no evidence for impaired lung functions or radiographic lung abnormalities associated with exposure to mineral wools.

The experimental evidence for the fibrogenic potential is limited. Three inhalation studies showed that mineral wools did not produce pulmonary fibrosis in rats or hamsters following chronic inhalation exposure (Wagner et al., 1984; Le Bouffant et al., 1984; Smith et al., 1986), whereas chrysotile asbestos used as the positive control induced extensive lung fibrosis in the

exposed rats (Wagner et al., 1984). However, the results of a limited inhalation study involving two rats which showed a production of focal fibrosis following chronic exposure to rock wool, provided suggestive evidence of a fibrogenic potential (Johnson and Wagner, 1980). The fibrogenic concern is further supported by the findings that respirable fractions of mineral wools were cytotoxic to cells in culture (Brown et al., 1979b; Davies, 1980). However, mineral wools were less cytotoxic than crocidolite asbestos.

II.2.5. Recommendations

Since the effects of mineral wools have not been adequately tested in animals, additional long-term inhalation studies on mineral wools are recommended. Additional epidemiological studies are also needed in order to fully assess the health hazard potential of mineral wools in humans.

II.3. Ceramic Fibers

Ceramic aluminum silicate glasses are vitreous substances, made by melting kaolin clay or a mixture of alumina and silica, and then blowing the melt to form the fibers. Alumina and zirconia fibers are monocrystalline substances, composed mainly of aluminum oxide and zirconium oxide, respectively. Ceramic fibers have high temperature resistance, and for that reason are often referred to as refractory ceramic fibers. Ceramic products are primarily used in industrial settings as high temperature insulation. The desired range of fiber diameters for industrial

applications is 2-3.5 μm, but the diameters can range from less than 1 um to 12 μm. Thus, ceramic fibers are likely to generate respirable airborne fibers (NRC, 1984).

II.3.1. Fiber Deposition, Clearance, and Retention

Very limited information is available on the lung deposition, clearance and retention of ceramic fibers. The results of available studies indicate that only a small fraction of inhaled ceramic aluminosilicate fibers was deposited in the rat lung. Fibers deposited in the lung were predominantly short (<10 μm) and thin (<1 μm). Ceramic fibers were found to be cleared slowly from the rat lung. Like other fibrous materials, ceramic fibers are presumably cleared by macrophage uptake, transport to the lymphatic system and possibly by slow dissolution and fragmentation.

The pulmonary deposition of ceramic fibers has been studied in the rat. Rowani and Hammad (1984) exposed 19 albino male rats (nose-only) to a ceramic aluminium silicate dust cloud with an average concentration of 709 fibers/mL for 5 consecutive days. The airborne fibers had a median length of 3.7 μm and a median diameter of 0.53 μm. The pulmonary deposition of fibers was measured 5 days after the last day of exposure; this period was allowed for clearance of approximately 95 percent of fibers deposited in the ciliated airways. Fiber deposition of the entire lung was 6.7 percent. Fibers that were deposited in all lobes were predominantly thin (<1 μm) and short (<10 μm). Large diameter and long fibers which constituted only a small fraction

of total lung deposition, were found to be higher in the lobes with relatively short path length from the trachea to the terminal bronchioles (left lung and right apical lobe). However, the overall fiber size distributions in the various lobes were not significantly different.

In another study, Hammad (1984) studied the clearance of ceramic fibers from the rat lung. In this study, a group of 49 male rats were exposed by "nose-only" to ceramic fiber (aluminum silicate) dust clouds with a mean concentration of 300 fibers/mL for five consecutive days. The mean diameter and length of airborne fibers were 0.7 and 9.0 μm, respectively. Rats in groups of seven were sacrificed 5, 30, 90, 180, and 270 days after the last day of exposure. The clearance of ceramic fibers from lung tissues was determined by the percentage of initial fiber retention at day 5 after exposure. The results of this study indicate that ceramic fibers are cleared slowly from the rat lung. The fiber clearance during the 5-30 and 30-90 day periods had the same half-lives of 85 days. The fiber clearance for the 90-180 and 180-270 day periods was much slower, with half-lives of 157 and 196 days, respectively. After 270 days following exposure, about 25 percent of the ceramic fibers were still present in the lung tissue.

There is very limited information on the clearance mechanisms of ceramic fibers. In the rat, alumina fibers were detected in the alveolar macrophages and in the mediastinal lymph nodes following inhalation exposure. These results suggest that these fibers may be transported from the lung via macrophages into the

lymphatic system (Pigott et al., 1981). Furthermore, large number of fibers were found in the sternal and mesenteric lymph nodes following injection of alumina or zirconia fibers into the abdominal cavity of rats. These findings also indicate that these fibers can migrate from the peritoneum into the lymph nodes and can be subsequently removed by the lymphatic system (Pigott and Ishmael, 1981; Styles and Wilson, 1976).

Ceramic fibers may also be cleared by dissolution and fragmentation. Hammad et al. (1985) reported that considerable alterations in the elemental composition were found in ceramic fibers (aluminum silicate) recovered from the rat lung after a long period of 270 days following short-term exposure to the fibers via inhalation. Furthermore, Pigott et al. (1981) observed that alumina fibers retained in the rat lung showed a high degree of fragmentation. These *in vivo* observations are consistent with results obtained from an *in vitro* study which showed considerable dissolution of ceramic fibers in physiological saline solution. However, ceramic fibers are relatively more resistant to solvent attack in comparison with other man-made mineral fibers including fibrous glass and mineral wools (Leineweber, 1984).

II.3.2. Effects on Experimental Animals

The potential oncogenic and fibrogenic effects of ceramic fibers have been evaluated in several animal studies via inhalation and injection routes of exposure. The experimental

design and results of available studies are summarized in Table 3 (pages 234-239).

II.3.2.1. Oncogenicity

The oncogenicity of ceramic fibers in laboratory animals appears to vary considerably for different fiber types. In one inhalation study (Davis et al., 1984), an increased incidence of lung tumors was observed in rats after chronic exposure to ceramic aluminum silicate glass. In another inhalation study, no lung tumors were produced in rats, but a single case of malignant mesothelioma was found in hamsters (Smith et al., 1986). Aluminum silicate fibers also produced mesothelioma in rats and hamsters by intrapleural or intraperitoneal injection (Davis et al., 1984; Smith et al., 1986; Wagner et al., 1973; Pott et al., 1987). In contrast, refractory alumina oxide and zirconia oxide fibers did not induce tumors in rats by either inhalation or intrapleural implantation (Pigott et al., 1981; Stanton et al., 1981).

II.3.2.1.1. Inhalation studies

Davis et al. (1984) studied the effects of long-term inhalation exposure to ceramic aluminum silicate glass in rats. In this study, a group of 48 SPF Wistar rats of the AF/HAN strain were exposed to the test dust cloud at a mass concentration of 8.4 mg/m^3 for 12 months. Approximately 95 fibers/mL were longer than 5 μm and less than 3 μm in diameter. Animals were sacrificed at 12, 18, and 36 months. The survival of treated and

control groups was similar. Eight exposed animals developed pulmonary neoplasms (1 adenoma, 3 carcinomas, 4 malignant histiocytomas). It should be noted that the pattern of tumor development appeared to be different than that for asbestos exposure since malignant histiocytomas have not generally been associated with asbestos exposure. One case of peritoneal mesothelioma was also observed in the exposed group. No pulmonary tumors of any types were found in the 40 unexposed control animals.

Smith et al. (1986) also studied the long-term health effects of refractory ceramic fibers in rats and hamsters. Male Syrian hamsters and female Osborne-Mendel rats were exposed "nose-only" to dust cloud of refractory ceramic fibers at a mass concentration of 12 mg/m^3 (equivalent to 200 fibers/mL) for 24 months. Approximately 83 percent of the exposure aerosol was greater than 10 μm long and 86 percent less than 2 μm in diameter (69 percent >10 μm long and <2 μm in diameter). The ceramic fiber type was not specified in the report but it was presumed to be aluminum silicate fibers (Kaolin) similar to the type that was tested by Davis et al. (1984).

In contrast to the test results of the study by Davis et al. (1984), no pulmonary neoplasms were observed in the exposed rats (0/55) in this study. None of the exposed hamsters developed primary lung tumors (0/70) but malignant mesothelioma was found in one hamster (1/70). This finding was not statistically significant although the possibility that the tumor was associated with ceramic fiber exposure could not be ruled out, since positive tumor responses have been observed in rats and

hamsters by the injection routes of exposure. With the exception of one bronchoalveolar tumor in a sham control hamster (1/58), none of the other sham controls or unexposed control animals developed pulmonary or pleural tumors. Thus, under the experimental conditions of this study, there was no evidence of carcinogenicity in rats exposed to refractory ceramic fibers and there was only suggestive evidence of carcinogenicity in exposed hamsters. It should be noted that in this study UICC crocidolite asbestos only produced a low tumor incidence in rats (3/57; 1 mesothelioma, 2 bronchoalveolar tumors) and no tumor response in hamsters. It was suggested that the lack of significant tumorigenic effects of crocidolite asbestos observed in this study could be due to the use of relatively short fibers (95-97 percent <5 μm long).

Pigott et al. (1981) investigated the effects of chronic inhalation exposure to refractory alumina fibers in rats and reported that they were not carcinogenic. Groups of 50 albino Wistar derived rats (25 of each sex) were exposed by inhalation to dust cloud of refractory alumina fibers (Saffil fibers), either as manufactured or in a thermally aged form at a mean respirable dust concentration of 2.18 and 2.45 mg/m^3, respectively, for 86 weeks, after which the animals were maintained to 85 percent mortality. No pulmonary tumors were reported in either animal groups exposed to manufactured Saffil fibers (0/42) or aged Saffil fibers (0/38). It should be pointed out that a major limitation of this study was the low levels of respirable dust clouds. Furthermore, both types of Saffil fibers had fairly

large diameters with median value of around 3 um. The tumor incidence found in positive control animals exposed to UICC chrysotile A asbestos at 4.54 mg/m^3 was comparable with those reported in other studies. Pulmonary neoplasms were found in 9 of 38 animals (5 adenomas, 3 squamous-cell carcinomas, 1 adenocarcinoma).

II.3.2.1.2. Intrapleural Injection/Implantation Studies

Wagner et al. (1973) found that the carcinogenic potency of ceramic aluminum silicate fibers, when administered via the intrapleural route, was considerably less than that of SFA chrysotile asbestos. Groups of 36 SPF Wistar rats (24 males and 12 females) were administered a single dose of 20 mg of ceramic or chrysotile fibers via intrapleural inoculation. The diameters of the tested ceramic fibers were between 0.5 and 1.0 um (lengths unspecified). Pleural mesotheliomas were observed in 3 of 31 rats treated with ceramic fibers. The survival time to the first mesothelioma was 743 days. In contrast, significantly higher incidences of mesothelioma were detected in rats treated with SFA chrysotile samples (21/32 with one sample and 23/36 in a second sample) as early as 325 days after injection. A concurrent vehicle control was not used.

Stanton et al. (1981) tested two ceramic glasses (presumably alumina and zirconia ceramic fibers) for carcinogenicity after intrapleural implantation (40 mg) into rats (50 female Osborne-Mendel rats/group). These were large diameter fibers, with microcrystalline aluminum oxide content >80 percent (glass 21),

and with microcrystalline zirconia oxide content greater than 90 percent (glass 22). Both ceramic fiber types were considered noncarcinogenic; 2/47 rats receiving glass 21 and 1/45 treated with glass 22 developed pleural neoplasms, compared to 3/491 untreated controls and 17/615 negative controls receiving noncarcinogenic pleural implants. As concluded by the investigators, the carcinogenicity of fibrous dust appeared to be related to fiber size; the lack of tumorigenic responses seen with the two ceramic glasses samples were probably due to the fact that they were composed of large diameter fibers.

II.3.2.1.3. Intraperitoneal Injection Studies

Davis et al. (1984) found that ceramic aluminum silicate glass was carcinogenic in rats by the intraperitoneal route. In this study, a group of 32 SPF Wistar rats of the AF/HAN strain (sex unspecified) received a single intraperitoneal injection of 25 mg of ceramic fibers. The injected fibers which were collected from the inhalation exposure system were predominantly short and thin (90 percent <3 μm long and <0.3 μm in diameter). Three animals developed peritoneal tumors (1 mesothelioma, 2 fibrosarcomas) approximately 850 days after injection. Negative control animals were not included in this study.

Smith et al. (1986) also reported that refractory ceramic fibers were carcinogenic in male Syrian hamsters and female Osborne-Mendel rats when injected as a single dose of 25 mg into the abdominal cavity of the animals. Fibers used for injections were collected airborne materials from the inhalation exposure

chambers with mean diameter of 1.8 um. The incidences of peritoneal mesothelioma in hamsters injected with refractory ceramic fibers were 13 percent (2/15) in one group and 24 percent (5/21) in a second group. The incidence of abdominal tumors in treated rats was 83 percent (19/23). On the other hand, 40 percent (8/25) of the hamsters and 80 percent (20/25) of the rats injected with crocidolite asbestos had abdominal mesotheliomas at their deaths. Negative saline controls and unmanipulated control animals had no tumors.

A recent study by Pott et al. (1987b) also showed that ceramic aluminum silicate fibers are highly carcinogenic in rats via the intraperitoneal route. Two ceramic fibrous samples were tested in this study. Suspensions of either Ceramic "Fiberfrax" dust (50 percent <8.3 μm in length; 50 percent <0.91 μm in diameter) or ceramic "MAN" (50 percent <6.9 μm in length; 50 percent <1.1 μm in diameter) were injected into the abdominal cavity of female Wistar rats at 5 weekly doses of 9 mg (a total dose of 45 mg) and 15 mg, (a total dose of 75 mg), respectively. High incidences of abdominal tumors were observed in both animal groups treated with ceramic Fiberfrax (33/47) and ceramic MAN (11/54). Under similar experimental conditions, UICC chrysotile asbestos induced comparable incidences of peritoneal tumors in a dose-related manner but at considerably lower doses, (11/36, 21/34, 30/36 at 0.05, 0.25, 1.0 mg, respectively). The saline control group had one tumor (1/102).

II.3.2.1.4. Intratracheal Instillation Studies

Smith et al. (1986) also tested the carcinogenicity of refractory ceramic fibers in male Syrian hamsters and female Osborne-Mendel rats via intratracheal instillation of 2 mg of the test fiber (mean diameter of 1.8 μm) once a week for 5 weeks (a total dose of 10 mg). All animals were maintained for the duration of their lives. Six of the 22 rats had bronchoalveolar metaplasia but none of the rats (0/22) nor hamsters (0/25) instilled intratracheally with refractory ceramic fibers developed primary tumors. In contrast, 74 percent (20/27) of the hamsters and 8 percent (2/25) of the rats instilled intratracheally with crocidolite asbestos developed bronchoalveolar tumors.

II.3.2.2. Fibrogenicity

The experimental data on the fibrogenicity of ceramic aluminum silicate glass are limited but suggestive of a fibrogenic potential. This ceramic fiber type was shown in one limited study to cause mild interstitial pulmonary fibrosis in rats by inhalation exposure. In contrast, available evidence indicates that large diameter alumina and zirconia fibers do not appear to cause fibrosis in rats via inhalation or by injection.

II.3.2.2.1. Inhalation Studies

In an inhalation study by Davis et al. (1984), as discussed under "Oncogenicity", ceramic aluminum silicate glass was found to induce low levels of interstitial pulmonary fibrosis in rats. Large areas of alveolar proteinosis and small amounts of inter-

stitial fibrosis were detected in the lungs of rats killed at the end of the 12 months exposure period and 6 months later (at 18 months). The fibrotic lesions were more severe and extensive (0.2 - 14.5 percent of total lung area) in older rats killed at 32 months, while only two of the control animals had very small areas of interstitial fibrosis (<0.01 percent of total lung tissue area).

Smith et al. (1986), however, did not observe lung fibrosis induction in rats or hamsters chronically inhaling (nose-only) refractory ceramic fibers for 24 months. In contrast, the number of hamsters and rats in the crocidolite asbestos exposure group with fibrous pulmonary lesions was statistically higher than either that of chamber or unmanipulated control group.

As discussed under "Oncogenicity", Pigott et al. (1981) reported that inhalation of Saffil or "aged" Saffil fibers (refractory alumina fibers) did not produce pulmonary fibrosis in the rat. Pulmonary reactions to both forms of alumina fibers were confined to minimal alveolar epithelialization. On the other hand, moderate fibrosis was produced in most rats exposed to UICC chrysotile asbestos. These results are not surprising considering that the tested ceramic fibers were relatively thick and contained low respirable fractions. Furthermore, the exposure concentrations of alumina fibers were low and almost twofold less than that of chrysotile.

II.3.2.2.2. Intraperitoneal Injection Studies

In a study by Pigott and Ishmael (1981), groups of 24 SPF Wistar rats (12 of each sex) were each injected intraperitoneally with 20 mg of refractory alumina fibers, either of Saffil type A (15.5 x 2.75 μm) or type B (17 x 3.7 μm), or UICC chrysotile asbestos (7.7 x 1.1 μm). Rats which were treated with either alumina fibers showed mild chronic inflammatory reactions and deposition of small amounts of collagen in the abdominal tissues after 3, 6, and 12 months following treatment. There was no evidence of progressive peritoneal fibrosis in these animals. In contrast, marked fibrosis was observed in rats given chrysotile at 6 and 12 months, whereas animals which were injected with vehicle control (saline) showed normal findings.

Styles and Wilson (1976) tested the fibrogenic potential of Saffil alumina and Saffil zirconia fibers (median diameter of 3.6 and 2.5 μm, respectively) and found that under the experimental conditions, these fibers were not fibrogenic in the rat. Groups of 40 Wistar rats (20 of each sex) were each injected intraperitoneally with a single 20 mg dose of either test materials or UICC chrysotile asbestos in saline (mean diameter of 1.1 μm). Control animals received saline only. All animals were killed 6 months after injection. Considerable number of white nodules containing fibroblasts and mononuclear cells and small amounts of collagen were detected in the abdominal cavity of 36 rats which were dosed with either Saffil zirconia or Saffil alumina fibers. However, there was slightly more collagen in animals treated with alumina fibers than in those dosed with zirconia

fibers. In contrast, animals which were treated with chrysotile showed evidence of marked peritoneal fibrosis.

II.3.2.2.3. Intrathoracic Injection Studies

In a short-term study by Davis et al. (1970), guinea pigs (number, sex and strain unspecified) were each administered by intrathoracic injection with 25 mg of ceramic aluminum silicate glass (mean diameter of 2 μm; 50 percent <75 μm in length). After 6 weeks following treatment there was considerable formation of ferrunginous bodies and large granulomas consisting of macrophages, giant cells and fibroblasts, in treated animals. These pulmonary reactions appeared similar to those induced by asbestos reported in other previous studies. Induction of lung fibrosis by ceramic fibers was not observed in this study. This finding is certainly not unexpected since the observation period of this study was quite short. In general, fibrotic lesions are produced by most mineral fibers including asbestos only after a minimum of 6 months following treatment.

II.3.3. In Vitro Studies

II.3.3.1. Genotoxicity

There is no information available with regard to the genotoxicity of ceramic fibers.

II.3.3.2. Cytotoxicity

Available in vitro data indicate that fibrous ceramic aluminum silicate is not cytotoxic to macrophage-like cells

(P388D1) and lung fibroblasts (V79/4 cells) but is active toward the human alveolar tumor cell line (A549). The results of a single study showed that alumina and zirconia ceramic fibers are slightly toxic to macrophages.

A respirable sample of ceramic aluminum silicate fibers collected from the inhalation chambers by an elutriation process was not found to affect the viability of P388D1 cells following a 48 h treatment at 50 mg/mL of the dust (Gormley et al., 1985). In contrast, almost all asbestos samples tested showed a wide range of cytotoxicity in this assay.

In a subsequent study (Brown et al., 1986), the same group of investigators tested the same ceramic fiber sample in two nonphagocytic assays. In the assay using Chinese hamster lung fibroblast (V79/4) cell line, respirable fibrous ceramic aluminum silicate was found to be inactive; the ED_{50}, the concentration of dust causing a 50 percent reduction in the cloning efficiency of V79/4 cells was greater than 100 mg dust/mL. On the other hand, the same fiber sample displayed some activity in the assay using human alveolar type II lung tumor cell line, as measured by the induction of growth and giant cell formation at 25 and 50 mg dust/mL. Ceramic aluminum silicate fibers ranked in the mid range in this assay; it was slightly more active than some of the amphibole asbestos tested but much less active than several chrysotile samples.

Styles and Wilson (1976) reported that Saffil alumina and Saffil zirconia induced low cytotoxicity in rat peritoneal macrophages whereas UICC chrysotile asbestos was highly

cytotoxic. It should be noted that both types of ceramic fibers contained predominantly large fibers (2-6 μm in diameter) while UICC chrysotile asbestos contained thin fibers (median diameter of 1.1 μm). The experimental details were not provided in this report.

II.3.4. Assessment of Health Effects

Available experimental studies indicate that ceramic aluminum silicate glass is carcinogenic and weakly fibrogenic in animals whereas large diameter refractory aluminum oxide and zirconia oxide do not appear to be tumorigenic nor fibrogenic. Therefore, it is concluded that ceramic aluminum silicate fibers may present a health hazard to exposed humans. Because of the variable results seen in available animal studies, a comparison of the carcinogenicty between ceramic aluminum silicate and asbestos cannot yet be determined. Based on available information, it would appear that refactory zirconia oxide and alumina oxide fibers would not pose significant health hazard in humans. The discrepant results concerning the pathogenicity of various types of ceramic fibers may be a function of variation in fiber size distribution.

There are no epidemiological studies available on the health effects from exposure to ceramic fibers. Available experimental studies indicate that ceramic aluminum silicate glass is carcinogenic in laboratory animals. The results of a study by Davis et al. (1984) showed that chronic inhalation exposure to ceramic aluminum silicate glass produced an increased incidence

of lung tumors in rats. In addition, injection of these fibers into the pleural cavity or abdomen of rats or hamsters have resulted in the production of mesothelioma of the pleura or peritoneum, respectively (Wagner et al., 1973; Davis et al., 1984; Smith et al., 1986; Pott et al., 1987b). Thus, based on sufficient evidence of carcinogenicity of ceramic aluminum silicate glass in multiple experiments with different routes of administration, but in the absence of human data, this ceramic fiber type may be categorized as a probable human carcinogen (Category B2).

Experimental evidence of fibrogenicity of ceramic aluminum silicate fibers is limited. However, the results of one chronic inhalation study by Davis et al. (1984) which showed low levels of lung fibrosis in the exposed rats suggest a low fibrogenic potential for ceramic aluminum silicate fibers. This finding is supported by in vitro test data showing that ceramic aluminum silicate is active toward the human cell line A549 (Brown et al., 1986).

With regard to other ceramic fiber types, refractory alumina oxide and zirconia fibers did not cause tumor or fibrosis in rats via inhalation exposure (Pigott et al., 1981) or intracavitary injection (Stanton et al., 1981; Pigott and Ishamael, 1981; Styles and Wilson, 1976). Similarly, the cytotoxicity of these fibers is low (Styles and Wilson, 1976). On the basis of available information, refractory alumina oxide and refractory, zirconia fibers are not classifiable as to human carcinogenicity due to inadequate evidence of carcinogenicity in animals and no

human data (Category D). A lack of pathogenic effects of these fibers may be because test fibers were largely nonrespirable.

II.3.5. Recommendations

It is recommended that epidemiological studies of exposed workers be initiated. No additional animal tests are recommended at this time. A large-scale animal study sponsored by industry is being conducted at a private laboratory. The study is designed to subject rats and hamsters to common types of ceramic fibers for long periods by inhalation and injection methods.

III. Naturally-Occurring Mineral Fibers

III.1. Erionite

Erionite is a naturally-occurring mineral of the fibrous zeolite class. Erionite and other natural zeolites are crystalline minerals that contain alkaline metal and alkaline earth elements in a hydrated aluminium silicate structure. In the United States, fibrous erionite is found in several well-defined deposits in Arizona, Nevada, Oregon, and Utah, where it occurs as thin, pure beds with sedimentary tuft sequences or as outcrops in the desert valleys of the intermountain region (Rom et al., 1983). Fibrous erionite has also been identified in the volcanic tuft located in the central region of Turkey (Spurny, 1983b).

Erionite can occur as either single needles or in clusters. Erionite fibers are, on the average, shorter than asbestos fibers with a maximum length of approximately 50 μm. Fiber diameter generally ranges from 0.25 μm to 1.5 μm (Wright et

al., 1983) although fibers with diameters of 0.01 to 5 μm have been reported (Suzuki, 1982). Airborne erionite fibers are generally respirable.

Erionite is rarely mined and has very limited commercial uses, mainly as a molecular sieve in ion-exchange processes. Erionite was once, but is no longer, used as a catalyst in petroleum cracking. Instead, synthetic nonfibrous zeolites are used extensively for these types of applications (ICF, 1986).

III.1.1. Fiber Deposition, Clearance and Retention

Little is known about the lung deposition of erionite fibers. A number of epidemiological studies have reported the detection of erionite in the pleural and parenchymal tissues in some Turkish villagers exposed to low ambient erionite concentrations (Baris et al., 1978; Rohl et al., 1982; Sebastien et al., 1981). Most of the erionite fibers found in the lung tissues were uncoated, with a mean diameter of 0.3 um and maximum length of approximately 9 μm. Some ferrunginous bodies containing erionite fibers (zeolite bodies) which were similar to typical asbestos bodies were also detected (Sebastien et al., 1981). Experimentally, it was reported in an abstract that erionite fibers were well distributed in the rat lung soon after intratracheal instillation, followed by the development of macrophage and giant cell granulomatous reactions, and the production of ferrunginous bodies after 1 week (Moatamed et al., 1981). These findings taken together indicate that airborne erionite fibers can penetrate the lung and pleura and appear to

elicit early lung tissue responses similar to those induced by asbestos. There is no information available on the clearance of erionite.

III.1.2. Effects on Experimental Animals

Erionite has been evaluated in several studies for potential carcinogenic and fibrogenic effects in animals by several routes of exposure. Table 4 (pages 240-242) summarizes the experimental protocols and results of available studies on erionite.

III.1.2.1. Oncogenicity

Erionite from different geographical sources has been shown to be extremely carcinogenic in rats by inhalation, and in both rats and mice following injection. Erionite appears to be more potent in inducing mesothelioma than either crocidolite or chrysotile asbestos.

III.1.2.1.1. Inhalation Studies

Wagner et al. (1985) tested samples of natural erionite and synthetic nonfibrous erionite for carcinogenicity in Fischer 344 rats via inhalation. Groups of 20 male and 20 female rats were exposed to mean respirable dust concentrations of 10 mg/m^3 for 7 hours/day, 5 days/week for 12 months. The tested dusts included Oregon erionite (86 percent <0.4 μm in diameter; 92 percent <10 μm long) at 354 fibers/mL, synthetic nonfibrous Zeolite (>0.5 μm) of similar chemical composition to erionite at 1040 particles/mL, and UICC crocidolite asbestos (95 percent <0.4

μm in diameter; 86 percent <10 μm long) at 1,630 fibers/mL. Twelve rats in each group were sacrificed at 12 months to study dust accumulation. The remaining animals were observed until death. An extremely high incidence of pleural mesothelioma was induced in rats (27/28) exposed to Oregon erionite. The average induction time was 580 days. One mesothelioma (1/28) and one adenocarcinoma (1/28) occurred in the rats exposed to the synthetic nonfibrous zeolite. It is interesting to note that no mesotheliomas were produced in any of the positive control rats exposed to crocidolite, and only one squamous carcinoma of the lung was observed (1/28). Unexposed control rats had no tumors (0/28).

Johnson et al. (1984b) examined the histopathology and ultrastructure of seven pleural tumors that had been induced in rats by inhalation to Oregon erionite. The tumors were epithelial, fibrosarcomatous, or mixed epithelial/sarcomatous in form. However, the majority of tumors were of mixed features with either the fibrous or epithelial component being more prominent. In general, erionite-induced mesothelioma appeared to be morphologically similar to human mesothelioma and to experimentally induced mesothelioma in rats by inoculation of asbestos into the pleural or peritoneal cavity.

III.1.2.1.2. Intrapleural Injection Studies

Three intrapleural studies also document the oncogenicity of erionite in rats. In the study by Wagner et al. (1985), groups of 40 Fischer 344 rats (20 of each sex) were each inoculated

intrapleurally with 20 mg of Oregon erionite (75 percent <6 μm long; 92 percent <0.2 μm in diameter), Turkish (Karain) rock fiber (91 percent <0.2 μm in diameter; 86 percent <6 μm long), synthetic nonfibrous zeolite or chrysotile asbestos. The test dusts were suspended in saline. A negative control group received only saline. All of the rats treated with Oregon erionite developed mesothelioma (40/40 rats). Karain rock fiber induced 38 mesotheliomas (38/40) while only two mesothelioma (2/40) occurred with the nonfibrous zeolite, which had similar chemical composition as erionite. Chrysotile asbestos produced a total of 19 mesotheliomas and one mesothelioma was found in the negative control group. The mean survival times in the Oregon erionite group (390 days) and Karain rock fiber (435 days) were considerably shorter than that in the chrysotile group (678 days).

Maltoni et al. (1982a) also reported induction of pleural mesothelioma in rats by erionite by intrapleural injection. Groups of 40 Sprague-Dawley rats (20 of each sex) received a 25 mg dose of either erionite or crocidolite asbestos in water. The dimensions of tested fibers were not specified. Among the 40 rats treated with erionite, 10 animals died within 53 weeks, 9 with pleural mesotheliomas. No pleural tumors were found in the crocidolite group after 53 weeks. None of the vehicle controls developed tumors. Followup data on the study have not been published.

Preliminary results from intrapleural studies by Palekar and Coffin (1986) showed that erionite induced pleural mesothelioma

in a dose-related manner in Fischer 344 rats (25 animals/dose level). A dose response (0.5-32 mg) was obtained for two samples of erionite tested, erionite I (mean length of 2.2 um; mean width of 0.25 um) and erionite II (mean length of 1.4 um; mean diameter of 0.17 um). The tumor response of both erionite samples was much greater than that of chrysotile and crocidolite when the data were expressed as the mass or the number of fibers.

III.1.2.1.3. Intraperitoneal Injection Studies

The evidence of the carcinogenicity of erionite following intraperitoneal injection is demonstrated by three studies in mice. Suzuki (1982) studied the carcinogenicity of erionite after a single intraperitoneal injection in male Swiss albino mice. In the first experiment, a group of 12 mice were each treated with 10 mg of erionite (20 percent <1 um and 95 percent <8 um in length; 19 percent <0.1 um and 94 percent <1 um in diameter). Seven mice served as untreated controls. Six treated mice were sacrificed at 2-3 months after injection to determine early pathological lesions. One treated mouse died from intestinal obstruction due to severe peritoneal fibrosis. Among the remaining five animals which died between 8 and 15 months after treatment, two had malignant peritoneal tumors. No tumors were found in untreated control animals. In a second experiment, groups of five mice received either 10 or 30 mg of erionite. A positive control group that also consisted of 5 animals were treated with 10 mg of chrysotile asbestos. A negative control group of 6 animals remained untreated. Four of five mice in the

low dose erionite group and 2/5 in the chrysotile group developed malignant peritoneal tumors. All mice in the high dose erionite group died with intestinal obstructions due to adhesion of the intestine. Untreated control mice had no tumors.

Suzuki and Kohyama (1984) subsequently reported a high incidence of peritoneal tumors, mainly malignant mesotheliomas, in male Balb/c mice treated with a single intraperitoneal dose of erionite. Two samples of erionite were tested. A group of 50 mice were administered 10 mg of erionite I (90 percent <8 μm and 6 percent >9.5 μm in length; 85 percent <1 μm and 8.7 percent >1.4 μm in diameter) while groups of 20, 50 and 75 mice received a single dose of 0.5, 2, or 10 mg of erionite II (95 percent <8 μm and 4 percent >9.5 μm in length; 82 percent <0.5 μm and 100 percent <1 μm in diameter), respectively. In the animal group treated with erionite I, 21 of 42 (50 percent) had malignant peritoneal tumors. Of the three groups treated with erionite II, 6 of 18 (33 percent), 24 of 44 (55 percent) and 3 of 8 (37.5 percent) had malignant tumors, respectively. Animals treated with 2 mg of chrysotile had no tumors (0/22) while 6 of 32 (18 percent) animals treated with 20 mg of chrysotile developed malignant peritoneal tumors. The erionite-induced mesotheliomas were similar to those induced by chrysotile asbestos in gross appearance and histology. Saline controls and untreated controls had no tumors (0/118 and 0/37, respectively).

Ozesmi et al. (1985) also tested dust from the village of Karain (Turkey) containing both fibrous and nonfibrous erionite for induction of tumors in Swiss albino mice using the

intraperitoneal route. Groups of Swiss albino mice (37-98/group) received a 5, 10, 15, 20, 30 or 40 mg dose of Karain dust in saline and were followed until death (up to 32 months). Mesothelioma developed in 41/321 of the dosed mice, malignant lymphomas in 31 and both lymphomas and mesothelioma in 11 animals, within 9 to 32 months after injection of dust. The incidence of tumors did not appear to be dose-related. Three mesotheliomas and one lymphoma occurred in 55 saline controls.

III.1.2.2. Fibrogenicity

There is no information available on the ability of erionite to induce fibrotic disease in animals by inhalation. However, erionite has been shown to cause fibrogenic effects in animals by the injection method.

III.1.2.2.1. Intrapleural Injection Studies

Erionite has been reported to produce a fibrogenic reaction when administered by intrapleural injection. In the study by Maltoni et al. (1982a), Sprague-Dawley rats were injected intrapleurally with 25 mg of erionite (dimension unspecified). Among the 40 treated animals, 10 died within 53 weeks, 9 with pleural mesotheliomas. Upon gross examination, the visceral, parietal, and diaphragmatic pleura appeared thickened and whitish. In addition, several hard, whitish or yellowish nodules from 2-10 mm in diameter were found scattered at different sites of the serosal surfaces. Deposits of erionite surrounded by granulomatous reaction were seen within the neoplastic tissue.

III.1.2.2.2. Intraperitoneal Injection Studies

Erionite has been shown to possess fibrogenic properties similar to asbestos following intraperitoneal injection. Severe fibrotic lesions were observed in the peritoneum of Swiss albino mice treated with single intraperitoneal injections of either 10 or 30 mg of erionite (Suzuki, 1982). Similar findings were observed in a followup study by Suzuki and Kohyama (1984), which reported that two different samples of erionite produced marked peritoneal fibrosis in mice by intraperitoneal injection, with a severity similar to that produced by chrysotile asbestos. The experimental details of these two studies are presented in the oncogenicity section.

III.1.3. In Vitro Studies

III.1.3.1. Genotoxicity

Available data indicate that erionite is genotoxic. The major genotoxic effects seen with erionite include DNA damage and repair, induction of cell transformation, clastogenicity and aneuploidy. Like asbestos fibers, erionite does not appear to cause detectable gene mutations.

Fibrous erionite (Oregon origin) was tested in C3H10T½ cells for transformation and unscheduled DNA synthesis (UDS) and in a human lung cell line (A549) for UDS (Poole et al., 1983b). The count median length was 1.7 μm and diameter was 0.2 μm. Erionite induced the appearance of transformed type III foci at concentrations >10 μg/mL (up to 30 μg/mL). These same investigators report that while erionite is not more cytotoxic

than asbestos, in another article they demonstrate that crocidolite and amosite asbestos do not transform 10T½ cells (Poole et al., 1983a).

Erionite was examined by two methods for UDS, the autoradiographic method with 10T½ cells and the liquid scintillation method with 10T½ and A549 cells (Poole et al., 1983b). With the autoradiographic method, erionite induced a significant increase in UDS over controls at 100, 150 and 200 µg/mL. However, at higher concentrations (250 and 500 µg/mL), no increases were seen, suggesting a cytopathic effect not measured in this study as no apparent cytotoxicity was noted. UDS was induced at all concentrations (50, 100, and 200 µg/mL) in both cell types with the scintillation method. The authors favored this method for use with fibers for two reasons: 1) fibers can obscure the nucleus in the autoradiographic method and therefore not allow a truly random selection of nuclei for counting; and 2) the scintillation method allows a larger number of cells to be assayed. This may reduce variability seen in autoradiographic UDS as cells do not receive a homogenous exposure to similar fiber lengths and diameters. Overall, erionite produced a significant level of UDS in two different cultured cell types indicating induced DNA damage and repair.

Palekar et al. (1987) reported that erionite fibers were at least as effective as, if not more than, asbestos in producing aneuploidy in exposed V79-4 cells. A comparable or lesser clastogenic effect than asbestos was also noted. A significant

reduction in diploid cells and a parallel increase over controls in aneuploid and polyploid cells were observed in cultures treated with erionite at all concentration levels ranging from 10-100 μg/mL, whereas in UICC crocidolite and UICC chrysotile-treated cultures, significant increases in aneuploidy were observed at all exposure levels except the low concentration, 10 μg/mL. When the effects were compared on the basis of number of fibers per dose, fewer erionite fibers than those of crocidolite and chrysotile were required to produce similar aneuploidy. Erionite treatment at 100 μg/mL also produced chromatid aberrations but the clastogenic effect of erionite was comparable to that of crocidolite, but weaker than that of chrysotile asbestos.

Further evidence that erionite fibers are clastogens and are capable of altering the ploidy of cultured cells is provided by Kelsey et al. (1986). These investigators studied the cytogenetic effects of Oregon erionite and crocidolite asbestos in Chinese hamster ovary (CHO) fibroblasts. Treatment with erionite at concentrations of 5-50 μg/mL induced a slight but significant elevation in sister chromatid exchanges (SCE) in cultures of synchronous CHO cells, while crocidolite at the same concentrations failed to significantly increase the frequency of SCE. However, both fiber types induced a low level of chromosomal aberrations in CHO cells. In addition, an increase in the relative percent of tetraploid CHO cells after treatment with either erionite or crocidolite was observed.

Kelsey et al. (1986) also tested the mutagenicity of erionite and crocidolite fibers in a human lymphoblastoid cell line (TK6) at either the HGPRT (hypoxanthine guanine phosphoribosyl transferase) or thymidine kinase loci. Both erionite and crocidolite were negative in these assays.

III.1.3.2. Cytotoxicity

Experimental evidence has demonstrated that erionite is hemolytic and cytotoxic to various cell types. In an abstract, Nadeau et al. (1983) reported that erionite caused hemolysis to rat erythrocytes. The hemolytic activity of erionite was lower than that of chrysotile but higher than that of amphibole asbestos (crocidolite, amosite, anthophyllite). It was also reported that erionite was cytotoxic to rat pulmonary alveolar macrophages in a dose-related manner. However, the experimental details including fiber dimensions and dose levels were not reported.

Palekar et al. (1985) studied the cytotoxicity of erionite and asbestos fibers in Chinese hamster ovary (CHO) and Chinese hamster lung V79-4 cell cultures at concentrations of 10-100 μg/mL during a 6-day exposure. In CHO cells, erionite and chrysotile asbestos were cytotoxic at 20 μg/mL and crocidolite asbestos was cytotoxic at 40 μg/mL. Similarly, V79 cytotoxicity was induced by erionite and chrysotile at 40 μg/mL while crocidolite was cytotoxic at 100 μg/mL.

Subsequent findings by Palekar and Coffin (1987) confirmed their previous results that erionite was cytotoxic to V79

cells. Erionite samples containing long erionite fibers (median length of 1.6 μm) exhibited similar cytotoxicity to UICC chrysotile asbestos by weight while shorter erionite fibers (median length of 0.99 μm) were less cytotoxic than chrysotile. However, both erionite samples were more cytotoxic to V79-4 cells than chrysotile when the cytotoxicity was expressed as a function of number of fibers. Both erionite samples were more cytotoxic to V79-4 cells than UICC crocidolite asbestos by either methods of data analysis.

III.1.4. Assessment of Health Effects

Erionite has been shown to be a potent carcinogen in animals and is potentially fibrogenic. Thus, there is sufficient evidence to conclude that erionite potentially poses a significant health hazard to the exposed humans. Based on experimental data, erionite appears to be at least as hazardous as asbestos.

III.1.4.1. Oncogenicity

Erionite may be categorized as a probable human carcinogen (Category B1) based on limited evidence of carcinogenicity from studies in humans and sufficient evidence from experimental studies.

Available epidemiological studies have shown that populations from several locations in South Central Turkey have a large excess incidence of malignant mesothelioma (Baris et al., 1978, 1981; Artvinli and Baris, 1979, 1982; Rohl et al., 1982).

There is limited evidence to suggest that erionite may be the major etiological factor. This is based on the findings that erionite was the major fibrous material present in the surroundings and in the air of these affected areas, and in the pleural and parenchymal tissues of individuals with pleural disease(s). However, since asbestos and other zeolite fibers were also found in environmental and tissues samples taken in one of the affected villages (Rohl et al., 1982; Baris et al., 1978; Boman et al., 1982), it is possible that asbestos and other fibrous agents could also be involved in the etiology of this malignant mesothelioma (Battelle, 1988).

Experimental studies have confirmed that erionite from Turkey and the United States is extremely carcinogenic in animals by several routes of exposure. In an inhalation study, a 96 percent incidence of malignant mesothelioma of the pleura was produced in rats following 1-year exposure to Oregon erionite (Wagner et al., 1985). Intrapleural inoculation of Oregon or Karain erionite also produced very high incidences of pleural mesothelioma (53-100 percent) in rats (Maltoni et al., 1982a; Wagner et al., 1985; Palekar and Coffin, 1986). In these studies, erionite caused more mesothelioma than either crocidolite or chrysotile asbestos by inhalation or intrapleural inoculation. Furthermore, the latency periods for mesothelioma induced by erionite were much shorter than those induced by asbestos. In mice, intraperitoneal injection of erionite resulted in the production of malignant mesotheliomas of the peritoneum at high yields, comparable to those induced by

asbestos (Suzuki, 1982; Suzuki and Kohyama, 1984; Ozesmi et al., 1985).

Positive findings from a few genotoxicity studies further support a carcinogenic concern. Erionite has been shown to cause DNA damage and repair (Poole et al., 1983b), cytogenetic changes including aneuploidy, chromosomal aberrations, sister chromatid exchanges (Kelsey et al., 1986; Palekar et al., 1987) and morphologic transformation of cells in culture (Poole et al., 1983b).

III.1.4.2. Fibrogenicity

In view of limited evidence from epidemiological studies and limited evidence from experimental studies, erionite is considered to be potentially fibrogenic.

Epidemiological evidence collected over the past several years from a limited geographical area in South Central Turkey where erionite was present indicated significant incidences of nonmalignant pleural diseases as well as malignant pleural mesothelioma. These nonmalignant chest diseases included calcified plaques, chronic pleural fibrosis, and pleural thickening (Baris et al., 1978, 1981; Artvinli and Baris, 1979, 1982). The evidence for erionite as the major etiological factor is considered to be limited because possible exposure to asbestos and other fibrous material cannot be excluded (Battelle, 1988).

No information is available on the ability of erionite to induce fibrotic diseases in animals by inhalation. However, erionite has been shown to cause fibrogenic effects in animals by

injection. Whitish or yellowish nodules and plaques associated with the visceral, parietal and diaphragmatic pleura, and pleural thickening were observed in rats given an intrapleural dose of erionite (Maltoni et al., 1982a). In mice, intraperitoneal injection of erionite produced severe peritoneal fibrosis which was intimately associated with the observed peritoneal tumors (Suzuki, 1982; Suzuki and Kohyama, 1984). The in vivo results are further supported by positive findings from in vitro studies showing that erionite is hemolytic and highly cytotoxic (Nadeau et al., 1983; Palekar et al., 1985; Palekar and Coffin, 1987).

III.1.5. Recommendations

Since erionite has been adequately tested in animals, no further animal testings are thought necessary. However, additional epidemiological studies should be conducted, if practical, to further evaluate the association between erionite environmental exposure and development of malignant and nonmalignant respiratory diseases.

III.2. Wollastonite

Wollastonite is an acicular or fibrous natural monocalcium silicate mineral. The largest deposits of wollastonite are located in the United States, Mexico and Finland. Wollastonite is widely used in ceramics, and as a substitute for asbestos in insulation, wallboard, and brake linings. Wollastonite exhibits similar heat resistance properties as asbestos but has lower tensile strength. Wollastonite fibers range from 1 to 10 μm in

diameter with an average diameter of 3.5 μm. The median fiber size of airborne wollastonite is 0.22 μm in diameter and 2.5 μm in length. Approximately 92-97 percent of total airborne fibers during mining and milling operations are considered respirable (ICF, 1986).

III.2.1. Fiber Deposition, Clearance and Retention

No information is available on the deposition, clearance, and retention of wollastonite.

III.2.2. Effects on Experimental Animals

Very few studies have been conducted to examine the oncogenic and fibrogenic effects of wollastonite in animals. Table 5 (page 243) summarizes the experimental protocols and test findings of these studies.

III.2.2.1. Oncogenicity

There is no information available regarding the oncogenicity of wollastonite in animals via inhalation exposure. However, wollastonite has been shown to be weakly tumorigenic by intrapleural implantation and nontumorigenic by the intraperitoneal route in rats.

III.2.2.1.1. Inhalation Studies

Recently, a chronic inhalation study in male Fischer 344 rats was conducted by the National Toxicology Program (NTP) to test for the oncogenicity of wollastonite. The experiment has

been completed but full results are not yet available. However, the authors reported that inhalation exposure to wollastonite produced no adverse effects on the animal survival (Adkins and McConnell, 1985) and no tumorigenic response (McConnell, 1988).

III.2.2.1.2. Intrapleural Implantation Studies

Stanton et al. (1981) showed that wollastonite was weakly carcinogenic in rats following intrapleural implantation with a 40 mg dose of particles. Four samples of wollastonite from a Canadian mine were tested. These fibers were relatively large and only one of these samples was completely fibrous. The incidences of pleural sarcoma observed at 2 years following treatment were: grade 1, 5/20; grade 2, 2/25; grade 3, 3/21; grade 4, 0/24. The tumor incidence in groups receiving grades 1 and 3 was statistically significantly higher ($p < 0.05$, Fisher exact test) than that of control animals implanted with noncarcinogenic materials (17/615).

III.2.2.1.3. Intraperitoneal Injection Studies

Pott et al. (1987b) recently reported that wollastonite was not tumorigenic in rats following intraperitoneal injection. In this study, female Wistar rats were injected intraperitoneally with 5 weekly 20 mg doses of wollastonite in saline. The test dust was obtained from India, with 50 percent of the fibers having diameters less than 1.1 μm and lengths less than 5.2 μm. The animals were observed for full life span. No peritoneal tumors (0/54) were found.

III.2.2.2. Fibrogenicity

There are no data available for evaluating the fibrogenic potential of wollastonite in laboratory animals. As mentioned above, a 2-year inhalation study of wollastonite has been completed to study the development of chronic respiratory disease in rats (Adkins and McConnell, 1985). According to a preliminary report, no evidence of pulmonary fibrosis was found in this study (McConnell, 1988) but data are not yet available for a full evaluation of the study. In the long-term intraperitoneal injection study by Pott et al. (1987b), wollastonite from India was found to cause a low degree of adhesions of abdominal organs as observed macroscopically in rats. However, it was not clear whether there were any developments of fibrosis in treated rats. Histological data are not yet available for a complete evaluation of this preliminary finding.

III.2.3. In Vitro Studies

III.2.3.1. Genotoxicity

There are no genotoxicity data available on wollastonite.

III.2.3.2. Cytotoxicity

Several in vitro studies have been conducted to compare the biological effects of wollastonite with asbestos with regard to the hemolytic activity to erythrocytes and cytotoxicity to macrophages. A number of investigators have shown that wollastonite was weakly to moderately hemolytic to human and rat erythrocytes while others have reported that wollastonite was

more or less unreactive to rat erythrocytes. Similarly, wollastonite was found to induce varying degrees of cytotoxicity in rat or rabbit alveolar macrophages, ranging from noncytotoxic to moderately cytotoxic. However, wollastonite was far less hemolytic and cytotoxic than asbestos. The conflicting *in vitro* results with wollastonite appeared to be related to different experimental conditions and the nature of the materials tested, especially particle morphology and size distribution.

III.2.3.2.1. Erythrocytes

Skaug and Gylseth (1983) tested the hemolytic activity of two samples of naturally-occurring wollastonite and three samples of synthetic nonfibrous calcium silicate in human red blood cells. Both wollastonite samples (one fibrous and the other mostly nonfibrous) were found to be weakly hemolytic while synthetic compounds were far more reactive. The particle size distribution of the tested fibers and dusts were not specified.

Hefner and Gehring (1975) studied the hemolytic activity of two wollastonite samples using rat erythrocytes. One sample was fibrous with a mean fiber length of 200 μm and the other sample was nonfibrous with a mean particle size of 4 μm. Both wollastonite samples were hemolytic but the large nonrespirable 200 μm wollastonite (fiber diameter not specified) had a much slower rate of hemolysis.

Similar results were obtained in a study by Potts et al. (1978) who studied the ability of two wollastonite samples to induce hemolysis in rat red blood cells. Large particle

wollastonite (200 μm) induced weak hemolytic activity whereas small particle wollastonite (6.73 μm) was moderately hemolytic. In contrast, chrysotile asbestos was found to be strongly hemolytic in this *in vitro* system.

In an abstract, Vallyathan et al. (1984) also reported that wollastonite is moderately hemolytic. Wollastonite fibers tested had lengths less than 10 μm. The hemolytic effects of chrysotile (<21 μm), amosite (41 μm), and crocidolite (<10 μm) were more pronounced than that of wollastonite. The source of red blood cells used in this assay was not specified.

In constrast, Nadeau et al. (1983) reported in an abstract that wollastonite was more or less unreactive in hemolytic assays using rat erythrocytes, whereas crocidolite asbestos was highly hemolytic followed by anthophyllite, amosite, and crocidolite. No other experimental details were provided in this report for evaluation.

III.2.3.2.2. Macrophages

Pailes et al. (1984) found that wollastonite had no cytotoxic activity in rabbit alveolar macrophages whereas chrysotile was strongly cytotoxic. Exposure of alveolar macrophages to wollastonite (fiber size distribution not specified) as much as 250 μg/mL did not induce lysosomal enzyme release (beta-glucuronidase, beta-galactosidase, acid phosphatase, and N-acetylglucosaminidase) or alter membrane integrity as measured by trypan blue exclusion and the release of the cytosolic enzyme, lactate dehydrogenase (LDH). On the other

hand, treatment of alveolar macrophages with as little as 25 µg/mL of chrysotile asbestos caused the release of lysosomal enzymes and decreased membrane integrity.

Similarly, Nadeau et al. (1983) reported in an abstract that wollastonite (fiber size distribution unspecified) was unreactive to rat pulmonary alveolar macrophages whereas dose-response relationships for cytotoxicity were observed with all asbestos samples. Cytotoxicity was evaluated by the release of cytoplasmic LDH and lysosomal alpha-galactosidase enzymes.

Warheit et al. (1984) also found that exposure of rat pulmonary macrophages to wollastonite did not affect cell viability or morphology but wollastonite exposure did result in a diminished phagocytic capacity of the cells. On the other hand, crocidolite asbestos affected both macrophage morphology and phagocytic activity without affecting macrophage viability. Wollastonite fibers were large and long compared to crocidolite fibers which were short and thin.

Vallyathan et al. (1984) reported in an abstract that the cytotoxicity of wollastonite appeared to be dependent on fiber length. Wollastonite containing fibers less than 10 um long was moderately cytotoxic in macrophage enzyme release studies (LDH, beta-glucuronidase, beta-N-acetylglucosaminidase) whereas shorter fibers (<5 µm) were only mildly cytotoxic. The cytotoxic effects of long-fibered chrysotile (<21 µm), crocodolite (<10 µm) and amosite (<41 µm) in alveolar macrophages were more pronounced than those of both wollastonite samples.

III.2.4. Assessment of Health Effects

Overall, there is some evidence to support a possible health hazard from exposure to wollastonite. However, results from available experimental studies indicate that wollastonite is much less biologically active than asbestos, suggesting that wollastonite may pose a lesser health hazard than asbestos.

III.2.4.1. Oncogenicity

Wollastonite may be classified as a possible human carcinogen (Category C) on the basis of limited experimental evidence of carcinogenicity and inadequate human data.

None of available epidemiological studies were designed to assess the risk of lung cancer or mesothelioma associated with wollastonite exposure. One case of retroperitoneal malignant mesothelioma has been reported in Finland in one worker who had been exposed to wollastonite for twenty years (Huuskonnen et al., 1983). However, no cause and effect relationship can be drawn based on a single case report. Preliminary information on an inhalation study of wallastonite in rats indicates the lack of tumorigenic response (McConnell, 1988). However, wollastonite has been shown in one study to produce weak tumorigenic response in rats via intrapleural implantation (Stanton et al., 1981) but does not induce tumors in rats via the intraperitoneal route (Pott et al., 1987b).

III.2.4.2. Fibrogenicity

Available data are inadequate to assess the fibrogenic potential of wollastonite. There were no reports available that

examined the fibrogenicity of wollastonite in animals. Limited epidemiological studies conducted to date on quarry workers in the U.S. (Shasby et al., 1977, 1979; Hanke et al., 1984) and Finland (Huuskonen et al. 1983, 1984) indicate a possible association between wollastonite exposure and some nonmalignant diseases such as impaired ventilatory capacity, mild fibrosis of the lung, pleural thickening, and chronic bronchitis. However, these studies do not provide conclusive evidence of nonmalignant respiratory disease following exposure since the sample size was small and exposure was relatively short (Battelle, 1988).

Nevertheless, available epidemiological findings do raise a concern for potential fibrogenicity of wollastonite, particularly in light of positive results from in vitro cytotoxicity assays, which are thought to be indicative of fibrogenic activity. Wollastonite has been shown to induce varying degrees of hemolytic and cytotoxic activity although it is far less active than asbestos (Skaug and Gylseth, 1983; Hefner and Gehring, 1975; Potts et al., 1978; Vallyathan et al., 1984). These in vitro findings suggest that wollastonite may be considerably less fibrogenic than asbestos.

III.2.5. Recommendations

In order to fully assess the health effect of wollastonite, additional epidemiological studies are needed. In addition, the results of the NTP inhalation bioassay should be evaluated.

III.3. Attapulgite

Attapulgite is a naturally-occurring sorptive and gelling clay made up of fibrous aluminum and magnesium silicate. Although attapulgite is mined commercially in several countries, the United States is a leading producer of attapulgite, the majority of which is mined in the areas of Quincy, FL and Attapulgus, GA. Attapulgite is used in a wide variety of applications as an absorbent and thickening agent, and to a lesser extent as a substitute for asbestos in friction products and other materials (NRC, 1984).

Attapulgite morphology can vary greatly depending on where the material is mined. The attapulgite in commercial use in the United States consists of short fibers (0.1-2.5 μm) with mean diameter of 0.07 μm (0.02 - 0.1 μm) (Zumwalde, 1977). French attapulgite fibers are also short (<1.2 μm) (Bignon et al., 1980) while attapulgite samples from Spain can either be long or short (Wagner, et al., 1987). All attapulgite fiber types are of respirable size.

III.3.1. Fiber Deposition, Translocation, and Clearance

There is very little information available on the deposition, translocation, and clearance of attapulgite. The results of two limited case studies suggest that attapulgite fibers are capable of penetrating into the alveolar spaces following inhalation, and that after ingestion, attapulgite can be transported to the kidney and excreted in the urine. There is also some experimental evidence suggesting that short attapulgite

fibers are readily cleared from the lung while longer attapulgite fibers appear to be retained longer in the lung.

Bignon et al. (1980) reported the findings of two case studies. In the first case, a high concentration of attapulgite fibers (42,000 fibers/mL) were found in lung washing fluid recovered by bronchoalveolar lavage from a 41-year-old patient with lung fibrosis, who had been exposed to attapulgite for 3 years during mining and processing of attapulgite. Mean length and diameter of the attapulgite fibers in lung washing fluids were 1.5 μm and 0.11 μm, respectively. In a second case, a very high concentration of attapulgite was found in the urine of a 60-year-old woman treated orally for 6 months with an attapulgite-containing drug at a fairly large dosage (6-9 g/day).

Wagner et al. (1987) examined the recovered dusts from the lungs of rats following 12 months exposure to two attapulgite dusts at 10 mg/m^3. Examination of both macerated lung tissue and ashed lung sections from animals exposed to short-fibered attapulgite (all fibers <2 μm long) from Lebrija (Spain) showed a complete absence of short fibers. On the other hand, in animals exposed to long-fibered attapulgite (palygorskite from Leicester, U.K.), fibers up to 25 μm in length and with diameters less than 0.2 μm were found in the macerated lung and ashed sections.

III.3.2. Effects on Experimental Animals

There is considerable information available on the effects of attapulgite in laboratory animals. The experimental protocols and results of available studies are summarized in Table 6 (pages

244-245). Attapulgite fibers from various geographical locations appear to have different pathogenic potential which may be related to differences in fiber size distribution.

III.3.2.1. Oncogenicity

A number of studies have been conducted to evaluate the oncogenic potential of attapulgite by different routes of administration. In a long-term inhalation study, attapulgite from Spain (Lebrija) which consisted of short fibers (all <2 μm) did not induce tumors in rats. Several injection studies also showed that short attapulgite fibers (<2 μm) from the United States (U.S.), Spain (Lebrija), and France did not cause mesothelioma in rats by the intrapleural or intraperitoneal routes of administration. Tumors were also not observed in mice following lifetime feeding with short-fibered attapulgite. In contrast, inhalation exposure to attapulgite (also known as palygorskite) from the United Kingdom (Leicester) which contained a considerable number of long-fibers (>6 μm) resulted in low incidences of lung tumors and mesothelioma in rats. In addition, long-fibered attapulgite samples from the U.K. (Leicester), Spain (Torrejon) and an unknown source produced high incidences of mesothelioma in rats via intrapleural or intraperitoneal injection.

III.3.2.1.1 Inhalation Studies

In a recent report, Wagner et al. (1987) provided the completed findings of a series of inhalation experiments on

attapulgite and asbestos. Two samples of attapulgite were tested, Lebrija attapulgite from Southern Spain, and palygorskite (synonymous to attapulgite) from the U.K. (Leicester). All fibers in the Lebrija attapulgite sample were less than 2 μm in length whereas the palygorskite sample from Leicester also consisted of long, thin fibers (18 percent ≥6 μm in length and <2.0 μm in diameter). In this study, groups of 40 SPF Fischer rats (20 of each sex) were exposed to a dust cloud of either attapulgite sample or UICC crocidolite asbestos at 10 mg/m^3 for 6 hours a day, five days a week for up to 12 months. Four animals (2 of each sex) were sacrificed after 3, 6 and 12 months of exposure to assess the severity of pulmonary fibrosis. The remaining 28 animals were allowed to live out their normal life span.

No significant tumor response was found in the animal group exposed to short-fibered attapulgite from Lebrija. Among the 40 exposed animals, there was only one peritoneal mesothelioma and 3 bronchoalveolar hyperplasia (BAH). It should be noted that BAH is considered to be a reaction to an irritant and not a tumor. In contrast, there was some evidence of carcinogenicity for the long-fiber palygorskite sample from Leicester in rats. Three mesotheliomas (two pleural and one peritoneal), one malignant alveolar tumor (MAT, accepted as an early carcinoma), two benign alveolar tumors (BAT) and 8 BAH (one BAH with MAT) were found among the 40 animals exposed to the palygorskite sample. A comparison of the carcinogenic potency between this attapulgite sample and asbestos could not be made based on the results of

this study alone because of the lack of significant tumorigenic response in the positive control group exposed to UICC crocidolite asbestos (1/40 adenocarcinoma, 2/40 BAH, 1/40 BAH with adenocarcinoma). No tumors were found in the unexposed controls (0/40) and the negative controls exposed to nonfibrous kaolin dust (0/40).

III.3.2.1.2. Intrapleural Injection Studies

Stanton et al. (1981) tested two samples of American attapulgite (Attapulgus, GA) for carcinogenicity in female Osborne-Mendel rats using an intrapleural implantation method. Both samples were composed of small-diameter short fibers. No excess of tumors was found in animals treated with 40 mg of either sample (2/29 for both samples) compared to untreated animals (3/491) and negative controls treated with noncarcinogenic implants (17/615).

Renier et al. (1987) also found no tumorigenic response with short, thin attapulgite from France in an oncogenic intrapleural bioassay. In this study, 20 mg of attapulgite fibers suspended in saline were injected into the pleural cavity of Sprague-Dawley rats (sex and number of animals not specified). The test fibers had a mean diameter of 0.06 um and a mean length of 0.77 μm. After 24 months of treatment, no pleural tumors were found. Positive control animals treated with UICC chrysotile or Canadian chrysotile asbestos had a 19 percent and 48 percent mesothelioma incidence, respectively. Vehicle control animals injected with saline had no tumors.

Wagner et al. (1982, 1987) conducted a series of injection studies with three samples of attapulgite and showed that two attapulgite samples which consisted of variable proportions of long fibers were highly carcinogenic by the intrapleural route whereas no excess tumors were produced by short attapulgite fibers. Forty SPF Fischer 344 rats, 20 of each sex, were inoculated with a single injection of one of the following dusts suspended in saline (dose unspecified) including Lebrija (Spain) attapulgite (<2 μm long), Torrejon (Spain) attapulgite (0.54 percent ≥ 6 μm long and <0.5 μm in diameter), palygorskite (18 percent ≥ 6 μm in length and <0.2 μm in diameter) from Leicester (U.K.), UICC crocidolite asbestos and chrysotile B asbestos. High incidences of pleural mesothelioma were found among animals treated with the palygorskite sample (30/32) and Torrejon attapulgite (14/40). These long-fibered attapulgite samples appeared to have comparable carcinogenic potency as UICC crocidolite and chrysotile B asbestos which induced 34/40 and 19/40 cases of mesothelioma, respectively. In the group treated with short-fibered attapulgite from Lebrija there were two mesotheliomas (2/40), one pleural and one peritoneal. The saline control group had one pleural mesothelioma (1/40).

III.3.2.1.3. Intraperitoneal Injection Studies

Pott et al. (1974) found that attapulgite from an unknown source which contained a large proportion of long fibers (30 percent >5 μm long) was tumorigenic when injected intraperitoneally in rats. In this study, a group of 40 Wistar

rats received three doses of 25 mg of attapulgite dusts at weekly intervals. A tumor incidence of 65 percent (peritoneal mesothelioma) was found for the attapulgite group, and the first tumor appeared at day 275. In animals treated with chrysotile asbestos, 30-67 percent had mesotheliomas. No peritoneal tumors were reported for saline control animals.

More recently Pott et al. (1985) reported that short, thin attapulgite fibers from France, Spain, and the United States induced no excess tumors in rats after intraperitoneal injection. However, no experimental details and results were available for evaluation.

III.3.2.1.4. Oral Studies

Brune and Deutsch-Wenzel (1983) reported that attapulgite was not tumorigenic in mice following lifespan feeding. In this study, groups of 60 male and 60 female NMRI mice were fed for 25 months with 1 percent or 3 percent of attapulgite (mean length <1 μm) admixed in a pelleted diet. Untreated animals served as controls. The animals were sacrificed at the end of the treatment period. Mortality rates were not influenced by the treatment of attapulgite. No toxic effects nor increase of tumors in any organs were observed.

III.3.2.2. Fibrogenicity

The results of a recent inhalation study indicates that long-fibered palygorskite (attapulgite) is fibrogenic in rats following long-term exposure. Short-fibered attapulgite, on the

other hand, appears to be nonfibrogenic under the same experimental conditions.

In the study by Wagner et al. (1987), 40 SPF Fischer 344 rats (20 of each sex) were exposed to either palygorskite fibrous dust from Leicester (U.K.) which contained a considerable proportion of long, thin fibers (18 percent $\geq$6 μm in length and <0.2 μm in diameter) or short attapulgite fibers (all <2 μm long) from Lebrija (Spain) at 10 mg/m^3 for 6 hours daily, five days a week for up to 12 months. A group of 40 positive control animals were exposed to UICC crocidolite asbestos at similar experimental conditions. Four animals (two of each sex) were sacrificed after 3, 6, 12 and 24 months to assess the severity of pulmonary fibrosis. The remaining animals were allowed to live out their normal life span and the oncogenic response was then evaluated upon sacrifice.

The mean fibrosis gradings evaluated from four animals killed at 3, 6 and 12 months after exposure to the long-fibered palygorskite were 3.0, 3.1, 4.0, respectively. Since a grading of 4.0 represents first signs of fibrosis, these results indicate that this palygorskite sample should be considered to be potentially fibrogenic in humans. It should be noted that a conclusive evaluation of the extent and progression of pulmonary fibrosis induced by this attapulgite sample in comparison to asbestos could not yet be made because (1) the gradings of pulmonary responses were not done for the palygorskite sample at 24 months; and (2) the low fibrogenic response in crocidolite asbestos exposed animals. In the majority of inhalation studies

with asbestos, extensive lung fibrosis with gradings greater than 4.0 is generally seen in exposed animals. In this study, however, the mean fibrosis gradings at 3, 6, 12 and 24 months for crocidolite exposed animals were only 4.1, 3.3, 3.1 and 3.8, respectively.

In contrast, the pulmonary response to short-fibered attapulgite from Spain was confined to the presence of dust-laden macrophages (grade 2) and early interstitial reaction (grade 3). The mean gradings of tissue response in the lungs of rats exposed to Lebrija attapulgite at 3, 6, 12, and 24 months were reported to be 3.1, 2.6, 3.2, and 3.2 respectively. Unexposed animals had normal lungs throughout the study period (gradings of 1.25-1.75).

III.3.3. In Vitro Studies

III.3.3.1. Genotoxicity

Little information is available for the genotoxicity of attapulgite. In one study, short attapulgite fibers were not found to induce unscheduled DNA synthesis (UDS) in primary cultures of rat hepatocytes.

Denizeau et al. (1985) tested the ability of short, thin attapulgite to induce DNA damage in primary rat hepatocyte cultures by measuring its capacity to induce UDS. Ninety-six percent of the attapulgite fiber had a diameter between 0.01 and 0.1 μm with an average length of 0.8 μm. The liquid scintillation UDS method was used. Attapulgite produced no UDS effect over controls at 10 μg/mL. The fiber did not induce

cytotoxicity, as assessed by the release of lactate dehydrogenase.

III.3.3.2. Cytotoxicity

In general, attapulgite from various sources was hemolytic to red blood cells and cytotoxic to macrophages; these *in vitro* effects were somewhat comparable to those induced by asbestos. Long-fibered attapulgite was found to induce cytotoxicity in nonmacrophage cells, whereas short attapulgite fibers were relatively inert. However, short attapulgite fibers were found to cause a nonsignificant increase in squamous metaplasia of hamster tracheal organ cultures.

III.3.3.2.1. Erythrocytes

A number of studies have shown that attapulgite was hemolytic to red blood cells from humans and animals. Jaurand and Bignon (1979) reported that palygorskite (attapulgite) was highly hemolytic. However, the information on the source and fiber size distribution of the attapulgite tested was not provided. Bignon et al. (1980) subsequently reported that long-fibered Spanish attapulgite (unspecified fiber length distribution) was more hemolytic to human red blood cells than UICC chrysotile asbestos. Three drugs containing short French attapulgite (mean length of 0.8 μm) were weakly hemolytic to human red blood cells. Other investigators found that attapulgite was as hemolytic as chrysotile asbestos to rat

erythrocytes (Nadeau et al., 1983) and sheep erythrocytes (Harvey et al., 1984).

III.3.3.2.2. Phagocytic cells

In an abstract, Nadeau et al. (1983) reported that attapulgite induced cytotoxicity in a dose-related manner to rat pulmonary alveolar macrophages similar to that induced by asbestos fibers. Cytotoxicity was evaluated by the release of cytoplasmic enzyme lactate dehydrogenase (LDH) and lysosomal enzyme alpha-galactosidase. Fiber dimensions and dose levels were not reported.

Similar findings were reported by Bignon et al. (1980) who studied the cytotoxicity of two drugs containing short attapulgite fibers (mean length of 0.6 um) in rabbit alveolar macrophages. Both drugs produced a 39-45 percent of LDH and 24-31 percent of beta-galactosidase release at 300 or 200 µg/mL.

Chamberlain et al. (1982) found that both short-fibered attapulgite and long-fibered attapulgite were cytotoxic to rat peritoneal macrophages. Treatment of cell culture with short-fibered attapulgite at 150 µg/mL caused a 58 percent of LDH release, while after treatment with long-fibered attapulgite there was a 29 percent of LDH release. Specific fiber dimensions were not provided in this study.

By using P388D1 macrophage-like cells, Harvey et al. (1984) found that attapulgite treatment at a relatively high concentration (1 mg/mL) for 4 hours caused considerable cytoxicity. However, attapulgite was slightly less cytotoxic

than UICC chrysotile A or Canadian chrysotile asbestos but was markedly more cytotoxic than crocidolite asbestos. This study did not provide the fiber size distribution of the attapulgite sample.

In contrast, Lipkin (1985) found no evidence of cytotoxicity of short-fibered French or American attapulgite in P388D1 macrophage-like cells. The maximum fiber length of both attapulgite samples was 1.2 and 1.6 μm, respectively. UICC amosite asbestos showed a dose-dependent cytotoxic effect on the macrophage system while both attapulgite samples had no effect at 10, 50 or 100 ug/mL, as measured by reduction in cell number over a 72-hour period.

III.3.3.2.3. Nonphagocytic cells

Chamberlain et al. (1982) reported that short fibered attapulgite at concentrations greater than 100 μg/mL induced no effect on colony formation of Chinese hamsters of V79-4 cells nor on the ability of human type II alveolar tumor (A549) cells to form giant cells, i.e., colonies containing more than 200 cells. Long-fibered attapulgite, on the other hand, reduced cloning efficiency of V79-4 cells by 50 percent at 52 μg/mL. UICC crocidolite asbestos was more potent than either attapulgite samples in inducing cytotoxic effects in both cell types.

Using I-407 human embryo cells, Reiss et al. (1980) found that short-fibered attapulgite from the United States (Attapulgus, GA) caused only minimal inhibition of colony

formation. At equal doses, amosite asbestos was considerably more cytotoxic than attapulgite.

III.3.3.2.3.4. Tracheal Organ Cultures

Woodworth et al. (1983) examined the ability of short attapulgite fibers to induce metaplastic changes in trachea mucosa of the Syrian hamster. Ninty four percent of attapulgite fibers were shorter than 1 μm long. The investigators reported that attapulgite treated explants underwent proliferative and metaplastic alterations. However, metaplastic changes were not statistically significant at 1, 4 or 16 mg/mL of attapulgite. In contrast, both long and short fibers of chrysotile asbestos induced a significant increase in metaplasia at low concentrations (1.0, 4.0 mg/mL).

III.3.4. Assessment of Health Effects

The toxicological properties of attapulgite may depend on fiber length. There is inadequate evidence of carcinogenicity and fibrogenicity of short attapulgite fibers (<2 um long) in humans and animals. Based on an apparent lack of significant effects from long-term animal studies via inhalation and by the intrapleural or intraperitoneal route, it would appear that short-fibered attapulgite from commercial deposits in the United States is less hazardous than asbestos. In contrast, there is sufficient experimental evidence of carcinogenicity and fibrogenicity for attapulgite samples containing long fibers (>5 μm in length). Available data bases, however, are not sufficient

to provide a definitive assessment with regard to the comparative pathogenicity between long-fibered attapulgite and asbestos. Fortunately, one of the samples (palygorskite from the U.K.) is of no commercial interest and the other sample is a Spanish product (Torrejon attapulgite) and is being used in the preparation of drilling mud in the exploration of oil deposits in the North Sea and Persian Gulf.

III.3.4.1. Oncogenicity

With regard to human carcinogenicity, short-fibered attapulgite (<2 μm in length) from the United States, France, and Spain, is not classifiable (Category D) because of inadequate evidence from epidemiological studies and insufficient evidence from animal studies. On the other hand, attapulgite containing long fibers (>5 μm in length) from Spain, and the U.K. may be categorized as a probable human carcinogen (Category B2) based on sufficient evidence of carcinogenicity in animal studies in the absence of human data.

The results of a single available cohort study (Waxweiler et al., 1985) provide inadequate evidence of human carcinogenicity of short-fibered attapulgite. This study examined the mortality trends among workers at one Georgia attapulgite operation. Lung cancer mortality in the total cohort was slightly elevated but a statistically significant excess of mortality due to lung cancer was observed among white workers while a deficit of risk was found in the nonwhite subgroup. The excess risk among white employees could have been related to exposure to attapulgite

because there was an increased risk among employees with presumed highest exposure levels, and an increased risk among those with longer duration of employment and time since first exposure. However, the relatively small size of the cohort and several other limitations such as the inability to confirm the completeness of the cohort, limit the conclusions that can be made about this study (Battelle, 1988).

Experimental studies indicate that short-fibered attapulgite fibers (<2 μm long) did not induce tumors in rats following chronic inhalation (Wagner et al., 1987) or by the intrapleural route (Stanton et al., 1981; Renier et al., 1987; Wagner et al., 1987), or intraperitoneal route (Pott et al., 1985). Short-fibered attapulgite also did not induce tumors in mice following lifetime feeding (Brune and Deutsch-Wenzel, 1983).

The negative results from a genotoxicity study provide supporting evidence for a lack of tumorigenic effect of short-fibered attapulgite. These fibers did not induce DNA damage in primary rat hepatocytes as reflected by a lack of an induction of unscheduled DNA synthesis (Denizeau et al., 1985).

In contrast, materials containing long attapulgite (>5 μm in length) have tested positive in long-term animal studies via various routes of exposure. Palygorskite from the U.K. (Leicester) was shown to induce low incidences of lung tumors and mesothelioma in rats following chronic inhalation and very high incidences of pleural mesothelioma by intrapleural injection (Wagner et al., 1987). Other attapulgite samples containing long fibers such as the one from a small deposit in Torrejon, Spain,

and an unknown source were also found to induce pleural mesothelioma by intrapleural injection (Wagner et al., 1987), and abdominal tumors by intraperitoneal injection (Pott et al., 1974), respectively. In all of these studies, the tumorigenic responses of long-fibered attapulgite were comparable to those induced by chrysotile and crocidolite asbestos.

III.3.4.2. Fibrogenicity

In view of inadequate evidence of fibrogenicity in humans and laboratory animals, there is an insufficient basis to support a health hazard concern for potential fibrogenic effects of short-fibered attapulgite. However, positive findings of several *in vitro* cytotoxicity studies on these fibers suggest a possibility of a fibrogenic hazard. As for the long-fibered attapulgite, there is sufficient evidence to conclude that prolonged exposure to the dust may cause the development of lung fibrosis in humans.

The results of three available studies reporting the effects of attapulgite exposure provide inadequate evidence of fibrogenicity of short-fibered attapulgite in humans (Battelle, 1988). One case of lung fibrosis in a worker who had been exposed to attapulgite for two years was reported, indicating a possible link between occupational exposure and fibrosis (Sors et al., 1979). However, the results of a morbidity study showed no consistent relationship between attapulgite exposure and respiratory symptoms (Gamble et al., 1985). Furthermore, Waxweiler et al. (1985) reported a deficit mortality risk for

nonmalignant respiratory diseases among attapulgite workers in a Georgia plant.

Experimentally, there are no studies available that examined the ability of short-fibered attapulgite in commercial use in the United States in inducing lung fibrosis in animals via inhalation. However, the results of a long-term inhalation study showed that attapulgite from Lebrija (Spain) which consisted of only short fibers did not induce fibrosis in rats (Wagner et al. 1987). Furthermore, none of the available injection studies with short-fibered attapulgite have reported the production of fibrotic lesions in rats via the intrapleural route (Stanton et al., 1981; Renier et al., 1987; Wagner et al., 1987) or intraperitoneal route (Pott et al., 1985). These results taken together suggest that short-fibered attapulgite is not likely to induce severe fibrogenic effects in humans. On the other hand, the fact that these fibers are hemolytic (Bignon et al., 1980; Jaurand and Bignon, 1979; Harvey et al., 1984; Nadeau et al., 1983) and cytotoxic to macrophages (Chamberlain et al., 1982; Bignon et al., 1980; Nadeau et al., 1983) suggests that a fibrogenic potential may exist.

With regard to long-fibered attapulgite, the results from a recent inhalation study (Wagner, et al., 1987) showed that the palygorskite from the U.K. (Leicester) that contained a considerable number of long particles caused lung fibrosis in rats following 12 months of exposure, comparable to that induced by crocidolite asbestos. These findings indicate that long-fibered attapulgite is potentially fibrogenic.

III.3.5. Recommendations

A long-term inhalation study in animals should be conducted to fully assess the chronic toxicity and oncogenic effects of attapulgite from a United States commercial deposit. Additional epidemiological studies should also be performed to further determine the health effects of attapulgite in humans.

IV. Synthetic Fibers

IV.1. Aramid Fibers

These synthetic fibers are formed from aromatic polyamides. Aramid fibers are characterized by high tensile strength, and chemical and flame resistance. They are used as replacements for asbestos in a number of applications such as insulation, flame barriers, thermal protective clothing and friction products.

There are two major types of aramid fibers produced in the United States, Kevlar® and Nomex®. Para-aramid Kevlar® is produced as continuous filament yarn, staple fiber (38-100 mm), short fiber (6-12 mm) or pulp (2-4 mm), with a nominal diameter of 12 um. Thus, Kevlar® fibers generally tend to fall outside of the respirable range. However, Kevlar® pulp, which is frequently used to replace asbestos, has fine curled or tangled fibrils (<1 um in diameter) attached to the surface of the core fiber, and these fibrils may break off and potentially become airborne upon abrasion during the manufacturing processes. Nomex® is manufactured as continuous filament, staple fiber and short fiber, with a nominal diameter of 12 um. However, unlike

Kevlar®, Nomex® does not have the tendency to form fine fibrils and therefore is not respirable (ICF, 1986).

IV.1.1. Fiber Deposition and Clearance

Available information on the pulmonary deposition and clearance of aramid fibers is very limited. Results of intratracheal instillation and inhalation studies indicate that the deposition of aramid fibers is dependent on fiber dimension and is dose-related. Short aramid fibers are mostly phagocytized by alveolar macrophages which are cleared from the lung via transport to the lymph nodes.

Reinhart (1980) reported that following intratracheal instillation (25 mg of Kevlar® polymer dust), large para-aramid fibers (100-150 μm in diameter) remained in the terminal bronchioles whereas small particulates (approximately 5 μm) penetrated deep in the aleveolar region of the rat lung. Following inhalation exposure to 0.1-18 mg/m^3 of ultrafine Kevlar® fibrils for 2 weeks, fiber deposition and macrophage response in the rat lung were found to be dose-related (Lee et al., 1983). At high concentrations, fiber dust and fiber-laden macrophages (dust cells) accumulated in the respiratory bronchioles and alveolar region immediately following exposure. Some dust cells containing short fibers (<2 μm long) were found in the peribronchial lymphoid tissue or tracheobronchial lymph nodes by 6 months post exposure. These results suggest that short Kevlar® fibers are cleared via phagocytosis followed by transport to the lymphatic system.

IV.1.2. Effects on Experimental Animals

The pathogenic potential of aramid fibers has been investigated in a single long-term inhalation study and a number of short-term inhalation studies as well as injection studies. The experimental protocols and results of available studies are summarized in Table 7 (pages 246-247).

IV.1.2.1. Oncogenicity

It has been shown that chronic inhalation to a dust cloud of ultrafine para-aramid (Kevlar®) fibrils resulted in increased lung tumor formation in rats. In addition, intraperitoneal injection of para-aramid (Kevlar®) pulp or fibers caused low incidences of peritoneal tumors in rats.

IV.1.2.1.1. Inhalation Studies

The results of a long-term inhalation study which investigated the oncogenic potential of ultrafine Kevlar® fibrils in rats are summarized in an unpublished report by Reinhardt (1986). In this study, 5 groups of male and female Sprague-Dawley rats (100 of each sex per group) were exposed to dust clouds containing ultrafine Kevlar® fibrils (90 percent <1.5 μm in width; more than 75 percent less than 20 μm long) at targeted concentrations of 2.5, 25, 100, or 400 fibrils/mL. The means of weekly Kevlar® fibril counts during the exposure period were 2.4, 25.4, 112.2 or 435.5 fibrils/mL, respectively (approximately equivalent to 0.08, 0.32, 0.63 or 2.23 mg/m^3, respectively). Rats were exposed for two years (6 hours/day, 5 days/week) with

the exception of males and females in the highest dose group which were exposed for only one year and then maintained for one year without exposure. This was due to high mortality and apparent lung toxicity found following one year of exposure at the highest dose. By the end of first year of exposure, 34 males and 15 females in the high dose group were found dead or were sacrificed in extremis.

Lung tumors identified as cystic keratinizing squamous cell carcinomas were observed in rats exposed to the two highest dose levels; in the group exposed to 400 fibrils/mL, 1 of 36 males (3%) and 6/56 females (11%) developed lung tumors. At 100 fibrils/mL, none of the exposed males had tumors (0/68) but lung tumors were found in 4 of 69 females (6%). Squamous cell metaplasias were also observed in 6 females exposed to 100 fibrils/mL. Control animals had no lung tumors.

IV.1.2.1.2. Intraperitoneal Injection Studies

Two intraperitoneal injection studies have also been conducted to determine the oncogenic potential of Kevlar® in laboratory animals. Pott et al. (1987b) reported a low tumor yield with Kevlar® in rats. Female Wistar rats (8 weeks of age) were administered 4 weekly intraperitoneal doses of 5 mg of Kevlar® (50% <3.9 μm and 90% <11 μm long; 50% <0.47 μm and 90% <0.75 μm in diameter) suspended in saline. Surviving animals were sacrificed at 130 weeks after first treatment. Three of 53 animals were found to have peritoneal tumors. The tumor incidence in the vehicle controls was 1/102.

The results of another intraperitoneal study conducted by Davis (1987) also showed a low tumor yield in rats with Kevlar® pulp. Three groups of male AF/Han strain rats (3 months of age) received a single intraperitoneal injection of a preparation of disaggregated filaments of Kevlar® pulp suspended in phosphate buffered saline at either 25, 2.5, or 0.25 mg of the test material. By mass the bulk of the injected material consisted of aggregates of large fibers. However, a small proportion of the injected sample was composed of free fibrils which were within the respirable range (96 percent <1 μm and 56 percent <0.25 μm in diameter). A group of untreated animals were maintained as controls. There were no significant differences in the survival between the treated and untreated groups. More than 50 percent of animals survived for more than 800 days and the oldest survivors exceeding the age of three years. Two animals in the high dose group consisting of 32 animals which received 25 mg of Kevlar® developed peritoneal mesothelioma. Both mesotheliomas were typical of those induced by asbestos. No peritoneal mesotheliomas were found in the two low dose treated group (0/32 in the 2.5 mg and 0/48 in the 0.25 mg dose groups) and the untreated controls (0/48).

IV.1.2.2. Fibrogenicity

Results of two inhalation studies indicate that ultrafine para-aramid (Kevlar®) fibrils induced low fibrogenic activity in rats. Animal studies with commercial grade para-aramid (Kevlar®) by inhalation and intratracheal instillation and Nomex® by the

intratracheal route did not produce lung fibrosis in rats. However, injection of para-aramid (Kevlar®) pulp into the peritoneal cavity of rats produced strong tissue reactions and a minimal degree of fibrosis.

IV.1.2.2.1. Inhalation Studies

Lee et al. (1983) studied the pulmonary response of male Crl:CD rats exposed by inhalation to ultrafine Kevlar® fibrils at 0.1, 0.5, 3.0 or 18 mg/m^3 and commercial Kevlar® fiber at 18 mg/m^3 for 2 weeks. Fiber length distribution at different exposure concentrations of ultrafine Kevlar fibers showed 60-70 percent of fibers were 10-30 μm in length and less than 1 μm in diameter. Only 13 percent of airborne commercial Kevlar® fibers were respirable. Five rats from each group were sacrificed at the end of 2 weeks of exposure. Subsequently, 5 rats from each group at 0.1, 0.5, 3.0 mg/m^3 exposed to ultrafine Kevlar®, and 5 rats exposed to commercial Kevlar® at 18 mg/m^3 were killed at 2 weeks, 3 months and 6 months post exposure. Five rats at 18 mg/m^3 exposure were sacrificed at 4 and 14 days, 3 and 6 months after exposure. Control animals exposed to air alone were sacrificed at the same intervals as the exposed rats.

The pulmonary response in rats exposed to commercial Kevlar® at 18 mg/m^3 and ultrafine Kevlar® at 0.1, 0.5, and 3.0 mg/m^3 essentially satisfied biological criteria for nuisance dusts, i.e., they did not produce significant collagen formation or permanent alteration of basic lung structure, and tissue reactions were reversible. However, ultrafine Kevlar® fibrils at

concentrations of 18 mg/m^3 produced minimal collagen fiber formation in the alveolar duct region where dust particles accumulated. It should be noted that this study was an assessment of the effects of short-term exposure and the results alone cannot be extrapolated to assess long-term hazard.

Reinhardt (1986) also reported a low level of lung fibrosis in rats chronically exposed to ultrafine Kevlar® at 2.5, 25 or 100 fibers/mL for 104 weeks or 400 fibers/mL for 52 weeks. Lung lesions observed in male and female rats included alveolar type II cell hyperplasia, bronchoalveolar hyperplasia, collagen fiber granulomas, cholesterol-containing granulomas, and formation of ciliated columnar cells in the alveolar ducts (alveolar bronchiolarization). A dose-response trend was evident with fewer, less severe lesions occurring in groups exposed to 25 fibers/mL and the highest incidences and most severe lesions present in rats exposed to 400 fibers/mL. No pathological events were evident in rats exposed to 2.5 fibers/mL. Experimental details of the study are provided in Section IV.1.2.1.1.

IV.1.2.2.2. Intratracheal Instillation Study

Reinhardt (1980) reported that intratracheal instillation of Kevlar® polymer dust produced only a non-specific dust cell reaction in rats. Rats (strain, sex, and total number of animals unspecified) were treated intratracheally with 25 mg of the polymer dust which contained a low proportion of respirable fibrous particles (<1.5 μm in diameter and between 5-60 μm in length) and a large proportion of larger nonrespirable particles

ranging up to 150 μm in diameter. Rats were maintained without further treatment and were sacrificed at 2, 7 and 49 days and 3, 6, 12 and 21 months after treatment. A group of control rats received saline only. No differences in mortality rates, clinical observations and gross autopsy results were observed throughout the study. Initially there was a nonspecific inflammatory response in the rat lung which then subsided within one week. In later sacrifices, however, foreign body granulomas containing dust particles were found along with a negligible amount of collagen. All tissue responses to dust particles decreased with increasing recovery periods.

Similarly, Reinhardt (1980) reported that intratracheal instillation of fibrous dust of Nomex® did not show any progressive pulmonary fibrosis in rats. Rats (strain, sex, and total number not specified) were instilled intratracheally with 2.5 mg of Nomex® suspended in physiological saline. The test material contained circular, oblong or rod-shaped particles varying in size from 2-100 um in length and 2-30 μm in diameter. Groups of rats (number unspecified) were sacrificed at 2 and 7 days, 3 and 6 months, and 1 and 2 years following exposure. Initial transitory acute inflammation followed by foreign body granuloma formation was produced by larger nonrespirable particles (3-100 μm). Respirable particles (<10 μm) produced only negligible dust cell reaction similar to that seen with nuisance particulates. After 2 years post-exposure, lungs appeared normal. It should be pointed out that the actual data of both intratracheal instillation studies were not provided in the report.

IV.1.2.2.3. Intraperitoneal Injection Study

In a long-term intraperitoneal injection study by Davis (1987), Kevlar® pulp was found to cause a low level of peritoneal fibrosis in rats. Histological examination of peritoneal tissues from rats injected with 25 mg disaggregated Kevlar® pulp taken at varying time periods between one week and 9 months after injection showed the formation of cellular granulomas. These granulomas consisted of macrophages and fibroblasts and foreign giant cells. There was also a small amount of fibrosis with deposition of reticulin and collagen fibers.

IV.1.3. In Vitro Studies

IV.1.3.1. Genotoxicity

There is no information available on the genotoxicity of aramid fibers.

IV.1.3.2. Cytotoxicity

The results of an in vitro study by Dunnigan et al. (1984) indicate that short, thin aramid fibers are at least as cytotoxic as chrysotile asbestos to rat pulmonary alveolar macrophages. Short, thin aramid fibers were extracted from commercial grade Kevlar®. Ninty percent of the counted fibers were less than 5 μm long and smaller than 0.25 μm in diameter. Average fiber length and diameter were 2.72 μm and 0.138 μm, respectively. The test fibers were added to freshly harvested pulmonary alveolar macrophages or cultured macrophages obtained from adult male Long-Evans black hooded rats, at concentrations of 0, 25, 50,

100, or 200 μg/mL. After an 18-hour incubation period, cytotoxicity was assessed by measuring lactate dehydrogenase (LDH) and beta-galactosidase enzyme released into the incubation medium. Releases of LDH (10-55%) and beta-galactosidase (0-48%) from pulmonary alveolar macrophages were essentially identical when either aramid or chrysotile fibers were incubated with freshly harvested cells. With cultured macrophages, the cytotoxic response was even higher with aramid fibers than chrysotile.

IV.1.4 Assessment of Health Effects

There is sufficient experimental evidence to conclude that ultrafine para-aramid is potentially carcinogenic and fibrogenic. Due to limited comparative data bases, it is not possible at this time to definitively assess the pathogenicity of ultrafine para-aramid relative to that of asbestos. This material, however, does not pose a health risk to humans because it is not available in commerce. For the commercial grades of para-aramid fiber and pulp to which humans are exposed, there is limited experimental evidence suggesting that they may have a low oncogenic and fibrogenic potential. Thus, a possible health hazard exists from exposure to commercial grade para-aramid particularly the pulp form which may generate respirable airborne fine fibrils upon abraison.

Nomex® aramid which contains mainly nonrespirable fibers do not appear to pose a significant health hazard to humans.

IV.1.4.1. Oncogenicity

Ultrafine para-aramid may be classified as a probable human carcinogen (category B2) on the basis of sufficient evidence of para-aramid carcinogenicity in animal studies and in the absence of human data.

There is no information available on the oncogenicity of para-aramid fibers in humans. The results of an inhalation study indicate that ultrafine para-aramid (Kevlar®) is tumorigenic in rats via inhalation (Reinhardt, 1986). It was shown that chronic inhalation exposure to ultrafine Kevlar® resulted in the development of malignant lung tumors in female rats in a dose-related manner (100 and 400 fibers/mL). No injection data are available on ultrafine Kevlar® to further assess its relative carcinogenic potency in comparison with asbestos. However, the positive findings from the inhalation bioassay are further supported by the weak tumorigenic responses observed in rats treated intraperitoneally with commercial grade para-aramid (Kevlar®) fibers (Pott et al., 1987b) and pulp (Davis, 1987) which contained small numbers of thin fibrils.

In the absence of epidemiological data and based on the limited evidence of carcinogenicity in animals, commercial grade para-aramid may be classified as a possible human carcinogen (Category C). The limited evidence of carcinogenicity in laboratory animals is provided by the positive results of two intraperitoneal injection studies which showed that Kevlar® fiber and pulp induced low incidences of peritoneal tumors in rats (Pott et al., 1987b; Davis, 1987). The authors attributed the

low tumorigenic responses of aramid fibers and pulps to technical difficulty in administering the test fibers, since they tend to aggregrate and form large clumps.

No experimental studies were available that evaluated the oncogenic potential of Nomex® aramid fibers. Thus, Nomex® is not classifiable as to human carcinogenicity (category D) due to inadequate data in humans and animals.

IV.1.4.2. Fibrogenicity

There were no epidemiological studies available that examined the potential fibrogenicity of aramid fibers. Results of two inhalation studies showed that ultrafine para-aramid (Kevlar®) is weakly fibrogenic in rats. Minimal pulmonary fibrosis was induced in rats by 6 months following a 2-week inhalation exposure to high concentrations (18 mg/m^3) of ultrafine Kevlar® fibrils (Lee et al., 1983). Furthermore, dose-related pathological lung effects including alveolar type II hyperplasia, alveolar broncholarization, and collagenized fibrosis were also observed in rats following long-term inhalation exposure to ultrafine Kevlar® at 25, 100 and 400 fibrils/mL (equivalent to approximately 0.3, 0.6 and 2.23 mg/m^3, respectively (Reinhardt, 1986). The concern for the fibrogenic potential of ultrafine Kevlar® is further supported by findings of an *in vitro* study demonstrating that short, thin aramid fibers extracted from commercial grade Kevlar® are as cytotoxic as chrysotile asbestos to rat pulmonary macrophages (Dunnigan et al., 1984).

Lung fibrosis was not found for commercial Kevlar® in a short-term inhalation study in rats (Lee et al., 1983). However, a low fibrogenic effect has been demonstrated for commercial Kevlar® pulp via injection. Davis (1987) showed that inoculation of Kevlar® pulp into the abdominal cavity of rats resulted in a low level of peritoneal fibrosis. On the other hand, no fibrogenic effects were observed in rats instilled intratracheally with large nonrespirable Kevlar® or Nomex® aramid fibers (Reinhardt, 1980).

IV.1.5. Recommendations

In order to fully assess the potential health effects of para-aramid fibers or pulps, additional animal testings by inhalation or injection appear necessary. Because of a low health concern, no further testing is recommended for Nomex®.

IV.2. Carbon Fibers

Carbon fibers are synthetic fibers which are characterized by light weight, high tensile strength, flexibility, good electrical conductivity, thermal resistance, and chemical inertness (except to oxidation). Carbon fibers are mainly used as reinforcing materials in structural composites. They are currently not used as asbestos substitutes but may replace asbestos in thermal and electrical insulation, textiles, and friction products.

Carbon fibers (92 percent carbon by weight) are made by the carbonization (i.e., pyrolysis) of precursor polyacrylonitrile (PAN), rayon, or pitch fibers, with PAN-based carbon fibers being

the most common. Carbon fibers are manufactured as continuous or chopped fibers. The nominal diameter of carbon fibers range from 5-8 μm, which fall outside the respirable range. However, less than 25 percent of these fibers have diameters less than 3 μm and shorter than 80 μm, which are considered respirable. Furthermore, upon mechanical or thermal stress, carbon fibers may split longitudinally to finer respirable fibers (ICF, 1986).

IV.2.1. Fiber Deposition, Clearance and Retention

There is very limited information on the deposition and clearance of carbon fibers. Results of available inhalation studies in guinea pigs indicate that inhaled carbon fibers are capable of penetrating the alveoli. In the lung, nonfibrous carbon particles are phagocytized by alveolar macrophages while uncoated carbon fibers longer than 5 μm are found in the extracellular matrix. Carbon fibers appear to be cleared from the lung slowly as evidenced by the detection of uncoated carbon fibers and dust-laden macrophages in the lung even after 6 months to 2 years following exposure.

Holt and Horne (1978) exposed guinea pigs to high concentrations of a respirable dust cloud of carbon fibers for 7-24 hours. Most of the respirable dust (99 percent) was nonfibrous (370 particles/mL) and the airborne concentration of respirable fibers (1-2.5 μm in diameter and lengths up to 15 μm) was very low (2.9 fibers/mL). Examination of lung tissues revealed the presence of carbon particles to be intracellular in the cytoplasm of macrophages. The few carbon fibers found in the

lung that were longer than 5 um were still extracellular after 27 weeks post-exposure. These fibers were uncoated.

In a subsequent experiment, Holt (1982) exposed guinea pigs to dust of carbon fibers for 100 hours. The carbon dust was reported to be submicron in size and mainly nonfibrous. However, dust concentration and particle dimensions were not given. It was reported that phagocytosis of the dust particles commenced immediately one day after exposure but proceeded slowly, with the number of dust-laden macrophages continuing to increase up to 400 days post exposure. Macrophages containing dust began to decline after that but were still evident even after 2 years following exposure.

IV.2.2. Effects on Experimental Animals

A number of studies have been conducted to evaluate the oncogenic and fibrogenic potential of carbon fibers in laboratory animals via various routes of exposure. Table 8 (pages 248-250) summarizes the experimental protocols and findings of relevant studies on carbon fibers.

IV.2.2.1. Oncogenicity

No information is available on the oncogenicity of carbon fibers in animals via inhalation. Studies of carbon fibers in rats by intratracheal instillation, intraperitoneal injection, and intramuscular implantation have reported no tumorigenic response. Petroleum pitch-based continuous fibers were reported to be weakly oncogenic in mice by the dermal route, and

subcutaneous implantation of carbon fibers were reported to produce local sarcomas in rats. However, the results of these studies are questionable in view of inadequate reporting of the test results and/or the nature of the materials tested.

IV.2.2.1.1. Intratracheal Instillation Studies

A small intratracheal instillation study was conducted at the U.S. Air Force Aerospace Materials Research Laboratory by C. Olson. Carbon fibers were reduced to respirable size (20 percent <1 μm in diameter, with varying lengths) by partial oxidation at a high temperature and then injected intratracheally into the lungs of male Fischer rats. The rats were maintained over a 2-year period. As reported by Parnell (1987), no lung tumors were found at 200 days, 1 year or 2 years after treatment. This was a preliminary report and details of the study design (e.g., dosage, number of animals) and results were not given for full evaluation.

IV.2.2.1.2. Intraperitoneal Injection Studies

Parnell (1987) also reported that no tumor response was observed in male Fischer rats treated intraperitoneally with respirable carbon fibers. Mesotheliomas were not found in any of the treated rats at 200 days or 2 years post-treatment. No other experimental details were provided.

IV.2.2.1.3. Intramuscular Studies

Tayton et al. (1982) investigated the carcinogenic potential of intramuscular implantation of carbon fiber in strand and powdered forms in rats and found no signs of any malignant changes. In one experiment, a 1.5 μm length carbon fiber Graffil®, was inserted intramuscularly into the left gluteal muscle of 50 rats (unspecified sex and strain). A group of 50 control animals received an implant of black braided silk suture material. In a second experiment, groups of 10 rats had either a 5 μm length of carbon fiber (Graffil®) or the black silk (for control) tied around the periosteum of the left femur. In a third study, a group of 50 rats each received intragluteal injection of a suspension of powdered carbon fibers (unspecified particle size). All surviving animals were sacrificed at 18 months and morphological and histological examinations of the implants were performed. In all cases, minimal tissue reactions were observed and there was no evidence of malignant changes. It should be noted that the route of administration used in this study may not be relevant in providing information with regard to the potential of carbon fibers in inducing lung toxicity and carcinogenicity via inhalation.

IV.2.2.1.4. Subcutaneous Implantation Studies

In 1982, Maltoni et al. (1982b) reported initiation of testing of carbon fibers for oncogenicity in rats by subcutaneous implantation. Male and female Sprague-Dawley rats (40 per sex) were each subjected to subcutaneous implantation of a 2 cm

diameter disc containing 25 mg of carbon fibers (fiber dimension not specified). Animals were to be kept under observation until spontaneous death and a complete necropsy and histopathologic evaluation of the tissues were to be performed. Recently, Maltoni et al. (1987) reported in an abstract that the preliminary results of this study showed an induction of local sarcomas in carbon fiber treated rats, but no other details regarding the nature of the test material and test results were available for a full evaluation of the findings.

IV.2.2.1.5. Dermal Studies

DePass (1982) evaluated the potential of carbon fibers in inducing cancer of the skin in mice. Four types of carbon fibers were tested: (1) continuous filament (CF) pitch-based, (2) pitch-based carbon fiber mat (MAT), (3) polyacrylonitrile continuous fibers (PAN-based), and (4) oxidized PAN-based (PAN-oxidized) fibers. No tumorigenic response appeared to be elicited by PAN-based, MAT, or PAN-oxidized fibers. The CF pitched-based, however, were judged to produce a weak tumorigenic response.

Groups of 40 male C3H/HeJ mice each received a 25 µL application of the test material suspended in benzene (10% w/v), to the clipped skin of the back three times weekly until death. Each of the four fiber types was ground by mortar and pestle (particle size not specified). A group of negative controls received benzene only while positive control animals were treated with 0.1 percent methylcholanthrene in acetone. No skin tumors

were found in the groups treated with PAN-based fibers or vehicle control (benzene). In the CF pitch-based group, one papilloma (1/40) and one squamous cell carcinoma (1/40) of the skin at the application sites were found. Because of extremely low historical control incidence (0/285 in C3H/HeJ mice used as benzene controls at that laboratory), CF pitch-based fibers were considered to be marginally oncogenic under the conditions of the study. A very low incidence of various types of skin tumors were found in rats treated with MAT or PAN-oxidized fibers. However, MAT and PAN-oxidized fibers were considered to have questionable oncogenic potential because the tumors observed were distal to the application site.

IV.2.2.2. Fibrogenicity

A number of studies have been conducted to determine the fibrogenic potential of carbon fibers by various routes of exposure. With the exception of a report that carbon fiber is fibrogenic in rats via intratracheal instillation, other studies have not produced positive results. It should be noted, however, that available studies are of little value in evaluating the fibrogenicity of carbon fibers because of their limited scope or experimental design, and/or lack of information on the test material or test results.

IV.2.2.2.1. Inhalation Studies

Two inhalation studies were conducted in an attempt to evaluate the effects of short-term exposure to chopped PAN-

oxidized carbon fibers in guinea pigs. In the first study, Holt and Horne (1978) exposed 13 specific pathogen-free guinea pigs to carbon dusts for up to 104 hours. No pathological effects were found in the lung of exposed guinea pigs examined at 1-144 days post-exposure. It should be pointed out that 99 percent of the respirable dust generated was nonfibrous (370 particles/mL) and the levels of respirable carbon fibers were extremely low (2.9 fibers/mL). These fibers had diameters of 1.0-2.5 μm and lengths up to 15 μm.

In a subsequent study, Holt (1982) also reported no evidence of pathological changes in the lungs of guinea pigs exposed to submicron carbon dusts. Specific pathogen-free guinea pigs (2-9 per group) were exposed to carbon dust for 7-12 hours or 100 hours and single animals were killed at intervals after one to 720 days. The dust was reported to be submicron in size and mainly nonfibrous. Dimensional characterization of the dust was not provided, nor were dust concentrations reported. Exposed guinea pigs showed no lung fibrosis nor other pathology. However, it should be pointed out that these two studies only demonstrated that short-term inhalation of mostly nonfibrous respirable fragments of carbon fibers caused no adverse effects to guinea pigs.

Recently, Owen et al. (1986) conducted a subchronic inhalation toxicity study on PAN-based carbon fibers and reported no systemic toxicity nor progressive pulmonary dysfunction in the exposed rats. Four groups of 10 male Sprague-Dawley rats were exposed to carbon fibers for 6 hours daily, five days a week for

4, 8, 12, or 16 weeks and were sacrificed at the end of the exposure. A fifth group consisting of 20 animals were exposed for 16 weeks and were kept for 32 weeks post-exposure. The mean atmospheric concentration of carbon fibers was 20 mg/m^3, with a range of 16-23 mg/m^3. Carbon fibers had a mean diameter of 7 μm and lengths ranging from 20-60 μm. A similar number of control rats were exposed to air only and were sacrificed at similar schedules. One death occurred during the sixth week of exposure but was not considered related to treatment. Pulmonary function tests conducted prior to animal sacrifice did not show any significant or consistent changes in airway resistance. Histologic examination revealed no inflammatory or fibrogenic reaction in the lungs of exposed rats. A lack of an effect was not unexpected considering that the dust cloud contained mostly large nonrespirable fibers.

IV.2.2.2.2. Intratracheal Instillation

Troitskaya et al. (1984) reported findings on comparative fibrogenicity of carbon fibers and asbestos. Rats each received a single intratracheal administration of either chrysotile asbestos or one of two preparations of polyacrylonitrile-reinforced carbon fibers. The animals were examined 1-9 months after the administration of the dust. It was reported that chrysotile asbestos was several-fold more fibrogenic than either of the carbon fiber samples. No further details were provided.

In a study by Swensson (1979), as reported by Gross and Braun (1984), a mixture of carbon fibers (size distribution not

specified) and unspecified plastic was injected intratracheally into Sprague-Dawley rats (dose and number of animals not specified). The animals were maintained for eight months. Aside from an acute foreign body reaction during the first month after instillation, there was no indication of obstructive lung disease at 8 months as judged by analysis of collagen content in the lungs. No other details were given for conclusive evaluation.

Parnell (1987) recently reported that there was no evidence of any adverse effects in male Fischer 344 rats following intratracheal instillation of respirable carbon fibers. No treatment-related degenerative lesions of the lungs were observed in this long-term study. This was only a preliminary report and full results were not available for evaluation.

IV.2.2.2.3. Intraperitoneal Injection Studies

Styles and Wilson (1973) reported that carbon fiber was not fibrogenic in rats following intraperitoneal injection. A group of 6 male and 6 female SPF albino Wistar rats (200-250g) were injected intraperitoneally with carbon fibers at a dose of 50 mg/kg (10-15 mg per animal). Particle size of the test material ranged from 0.2-15 um in diameter. No pathological lesions were found at 1 and 3 months after treatment. On the other hand, rats injected with chrysotile asbestos developed diffuse fibrosis of the peritoneum after 1-3 months post-treatment. It should be noted that a major limitation of this study was the short observation period and the use of a small number of animals. It was also not clear as to whether the test carbon dust was in fibrous or nonfibrous form.

Parnell (1987) also reported negative findings with two samples of respirable carbon fibers following injection into the peritoneal cavity of male Fischer 344 rats. No treatment-related degenerative lesions were observed in either treated animal group at 200 days or 2 years after treatment. This was only a preliminary report and full results were not available for evaluation.

IV.2.2.2.4. Other Studies

Neugebauer et al. (1981) conducted a series of experiments to determine the reaction of tissues to carbon fibers. In this study, 50 mg of carbon fiber reinforced carbon (CFRC) fragments (diameter of 7 um and lengths between 20-100 um) were injected into the femoral medullary canal of 16 rabbits of the CHBB:CH strain. Tissue reactions were evaluated at 2 and 12 weeks post-treatment. A small amount of fibrosis and foreign body giant cell reactions were found in the medullary cavity. It was also reported that previous experiments in rats involving intravenous, intraperitoneal or intra-articular injections of carbon fiber particles (1-8 μm) also showed no evidence of tissue reaction. No additional details were available.

IV.2.3. In Vitro Studies

IV.2.3.1. Genotoxicity

Genotoxicity tests have been conducted on two types of carbon fibers. The carbon fibers are the acetone reconstituted benzene extracts of pitch-based carbon fibers and poly-

acrylonitrile (PAN)-based carbon fibers. Available data indicate that neither type of carbon fiber appears to cause gene mutations. However, pitch-based carbon fibers appear to be clastogenic while a clastogenic mechanism cannot be entirely ruled out for PAN-based carbon fibers.

Both carbon fibers were negative in the Salmonella/mammalian activation assay and the Chinese hamster ovary/hypoxanthine-guanine-phosphoribosyltransferase (CHO/HPRT) mutation assay (Litton Bionetics, 1980; Union Carbide, 1983a, 1983b, 1983c, 1983d). Pitch-based carbon fibers induced significant concentration dependent increases of sister chromatid exchanges (SCE) in CHO cells and unscheduled DNA synthesis (UDS) in primary rat hepatocytes (Union Carbide, 1983c). In the SCE assay, chromosomal aberrations were also noted for this carbon fiber. On the other hand, PAN-based carbon fiber did not induce significant increases in UDS in primary rat hepatocytes and the frequency of SCE in CHO cells. However, several types of chromosomal aberrations were observed in the SCE assay (Union Carbide, 1983d). These results are consistent with other fiber studies where mineral fibers do not appear to cause gene mutations, but are clastogenic.

IV.2.3.2. Cytotoxicity

Both positive and negative results were obtained from in vitro cytotoxicity studies on carbon fibers. Carbon fibers were reported in one study to be non-hemolytic to rabbit erythrocytes but highly cytotoxic to rabbit alveolar macrophages. However, in

another study, carbon fibers were found to cause no cytotoxic effects to either rat alveolar or peritoneal macrophages. Carbon fibers also did not affect rabbit lung fibroblast cultures. It would appear that the discrepancy in the observed in vitro biological activity of carbon fibers might be related to differences in fiber type and/or size distribution of the test materials.

IV.2.3.2.1. Erythrocytes

Richards and Hunt (1983) reported that carbon fibers had little or no hemolytic activity in rabbit erythrocytes. The test fibers were obtained by grinding carbon fiber cloth. Ninety percent of the fibers were less than 10 μm in length. Hemolysis was observed only at a relatively high dose compared to chrysotile asbestos. No other details were available.

IV.2.3.2.2. Phagocytic Cells

Richards and Hunt (1983) also reported that carbon fibers (90 percent <10 μm long) were highly cytotoxic to rabbit alveolar macrophages following one hour of incubation. Experimental details were not provided for full evaluation.

On the other hand, Styles and Wilson (1973) found that carbon dusts (0.2-15 μm in diameter) were not cytotoxic to rat peritoneal macrophages or alveolar macrophages. The test dusts were incubated with cells for 2 hours and cell viability was assessed at 0, 1 and 2 hours after addition of dust. Less than 2-5 percent of cells were killed following phagocytosis of carbon

dust. In contrast, chrysotile asbestos induced a high degree of cytotoxicity.

IV.2.3.2.3. Fibroblasts

Richards and Hunt (1983) tested the effect of ground carbon fiber cloth (90 percent <10 μm long) on rabbit lung fibroblast in culture. The amount of DNA and hydroxyproline levels in the culture were measured after 24 days of exposure. Treatment of fibroblast cultures with carbon fibers affected neither parameter.

IV.2.4. Assessment of Health Effects

Currently available data provide inadequate evidence of carcinogenicity and fibrogenicity for carbon fibers. However, based on suggestive evidence from a dermal study and positive clastogenic effects in genotoxicity tests with pitch-based carbon fibers, a weak oncogenic potential for certain types of respirable carbon fibers may exist. Overall, carbon fibers appear to pose a lower degree of health hazard compared to asbestos because they are less respirable, and less biologically active than asbestos as demonstrated in the few available comparative studies.

IV.2.4.1. Oncogenicity

Carbon fiber is not classifiable as a human carcinogen (Category D) based on inadequate evidence of carcinogenicity from animal studies and in the absence of human data.

No information is available on the potential development of respiratory malignant diseases in humans from exposure to carbon fibers. Moreover, there are no animal data on the oncogenic

potential of carbon fibers via inhalation. However, negative results have been reported in rats via intratracheal instillation (Parnell, 1987), intraperitoneal injection (Parnell, 1987), and intramuscular implantation (Tayton et al., 1982). The studies that reported positive results were those of a subcutaneous study with carbon fibers in which an increased production of local sarcomas were found in rats (Maltoni et al., 1987) and a lifetime skin painting study with pitch-based carbon fibers showing a nonstatistically significant increase of skin tumors in mice (DePass, 1982). The biological significance of these findings remains uncertain in light of the absence of particle size and morphology data, the weak tumorigenic response in the dermal study, the lack of data reported in the subcutaneous injection study as well as the questionable relevance of its method of administration to human exposure at the workplace. On the other hand, the positive clastogenic effects of benzene-extracts of pitch-based carbon fibers (Union Carbide, 1983c) tend to support an oncogenicity concern for this carbon fiber type. Additional data are needed to conclusively evaluate the oncogenicity of carbon fibers.

IV.2.4.2. Fibrogenicity

Available data are insufficient to evaluate the fibrogenic potential of carbon fibers. There is one single small cross-sectional study which showed no evidence of pathological effects in the lungs of workers in a PAN-based carbon fiber production plant, based on respiratory symptoms, spirometric and chest

radiographic data (Jones et al., 1982). It should be noted, however, that respirable fiber concentrations in this facility were low and that the duration of exposure to carbon fibers was relatively short. Thus, the results of this study do not provide conclusive evidence of a negative effect (Battelle, 1988).

With regard to experimental studies, there are no data available on the long-term effects of inhalation of respirable carbon fibers in animals. The results of a subchronic inhalation toxicity study showed no evidence of lung pathology in rats exposed to large diameter carbon fibers (Owen et al., 1986). In addition, several studies have reported that carbon fibers were not fibrogenic in rats via intratracheal instillation (Parnell, 1987; Swenson, 1979), intraperitoneal injection (Parnell, 1987; Styles and Wilson, 1973) or injection into the medullary cavity of femur bone (Neugebauer et al., 1981). On the other hand, it was reported that PAN-based carbon fibers induced lung fibrosis in rats following intratracheal instillation (Troitskaya et al., 1984). Most of these studies, however, are of little value for the evaluation of the fibrogenic potential of carbon fibers because of limited scope, lack of particle size and morphology data of the test materials, and/or no details available on study design and findings. Furthermore, both negative and positive findings have been reported regarding the in vitro cytotoxicity of carbon fibers (Styles and Wilson, 1973; Richards and Hunt, 1983). Thus, available animal and in vitro studies do not provide conclusive evidence for or against a fibrogenic effect for carbon fibers. They do suggest, however, that carbon fibers

at most have low fibrogenic potential, as supported by results of a few studies showing that chrysotile asbestos was more fibrogenic, hemolytic and cytotoxic than carbon dust fibers under the same experimental conditions (Troitskaya et al., 1984; Styles and Wilson, 1973; Richards and Hunt, 1983).

IV.2.5. Recommendations

A chronic inhalation toxicity study was recently conducted at a private laboratory. When results become available, they should be assessed to see if further study is warranted on this fiber.

IV.3. Polyolefin Fibers

Polyolefin fibers are manufactured from long-chain, synthetic polymers of ethylene, propylene or other olefin units. Approximately 95 percent of polyolefin fibers are made from polypropylene, while most of the rest is from polyethylene. Polyolefins are manufactured as monofilament yarn (greater than 153 μm in diameter), multifilament yarn (5-20 μm in diameter), tape and fibrillated film yarn (continuous sheet), spun-bonded fabric, staple fiber (chopped multifilament), synthetic pulp (5-40 μm in diameter, 2.5-3 mm long), and microfiber (1-5 μm in diameter). Applications of polyolefin fibers and pulp as substitutes for asbestos include roof sealant, asphalt solvent, caulks, joint cement, adhesive, textile compounds, filter, flooring felts, and roofing felts. With the possible exception of polyolefin microfibers, the likelihood that polyolefin fibers

and pulp generate airborne respirable fibers appears small since they generally fall outside the respirable range. Furthermore, it is unlikely that polyolefin fibers and pulp would split longitudinally to produce finer respirable fibers (ICF, 1986).

IV.3.1. Fiber Deposition, Clearance and Retention

There is no information available on the lung deposition, clearance and retention of polyolefin fibers.

IV.3.2. Effects on Experimental Animals

Very few studies have been conducted to determine the oncogenic and fibrogenic potential of polyolefin fibers in animals. Table 9 (page 251) summarizes the experimental protocols and results of available animal studies on polyolefin fibers.

IV.3.2.1. Oncogenicity

There is no information available on the oncogenic potential of polyolefin fibers in animals via inhalation. Preliminary results of an intraperitoneal injection study showed that polypropylene fibers induced a low incidence of peritoneal tumors in rats. In a limited intratracheal insufflation study, both polyethylene and polypropylene fibers were not tumorigenic in rats. Other forms of polyethylene and polypropylene including disc, film, rod fragment and powder produced local sarcomas in mice and rats following subcutaneous or intraperitoneal implantation (as reported in IARC, 1979).

IV.3.2.1.1. Intraperitoneal Injection Studies

Pott et al. (1987b) recently reported preliminary findings of a study of polypropylene fibers in rats via intraperitoneal injection. Female Wistar rats received 5 weekly injections of 10 mg of polypropylene fibers in saline. Ninety percent of the test fibers were less than 2.1 μm in diameter and less than 23 μm long. The animals were observed for full lifespan. Peritoneal tumors were found in 2/53 treated animals compared to 1/102 negative controls (saline). In contrast, animals treated with chrysotile asbestos showed a dose-dependent tumorigenic response at extremely low doses. The tumor incidences (identified as mesothelioma or sarcoma) in the chrysotile group were 11/36, 21/34, and 30/36 at a single dose of 0.05, 0.25, and 1.0 mg, respectively.

IV.3.2.1.2. Intratracheal Insufflation Studies

MB Research Laboratories (1980) conducted a long-term study of the effects of intratracheal insufflation of polyolefin fibers in rats. Groups of 40 male Long-Evans rats were administered a single dose of ozonized polyethylene SHFF, ozonized polypropylene SHFF, or HHF polypropylene. Control animals received vehicle only (Tween 60). All surviving animals were sacrificed at 21 months following administration of the test material. A number of deaths were observed in both control and treated groups. The cause of early deaths was attributed to dosing technique and that of later deaths to infectious diseases. Histologic examinations showed the development of lung granulomas in all treatment

groups. No lung tumors were found in any test groups. It should be pointed out that in the absence of available information on the characteristics of the test materials, specific dosages and methods of administration, this study is of little value for the evaluation of the oncogenic potential of polyolefin fibers.

IV.3.2.2. Fibrogenicity

There is no information available on the fibrogenic effects of polyolefin fibers in animals via inhalation. In a 3-month study, peritoneal fibrosis was not observed in rats following a single intraperitoneal dosing of polyethylene or polypropylene dusts. In a limited intratracheal study, lung fibrosis was not produced in rats treated with either polyethlyene or polypropylene fibers. However, preliminary results of a lifespan study in rats reported a strong degree of adhesions of the abdominal organs following intraperitoneal injection of polypropylene fibers.

IV.3.2.2.1. Intraperitoneal Injection Studies

In the study by Pott et al. (1987b) that was described in the oncogenicity section, a strong degree of adhesions of abdominal organs was observed macroscopically in rats treated with 5 weekly doses of 10 mg of polypropylene fibers (90 percent <2.1 μm in diameter; 90 percent <23 μm long). It was not clear whether there were any developments of fibrosis in treated rats. Histological data are not yet available for a full evaluation of this preliminary finding.

Styles and Wilson (1973) reported that polyethylene and polypropylene dusts were not fibrogenic in rats via the intraperitoneal route of exposure. In this study, groups of 6 male and 6 female albino Wistar rats (200-250g) were intraperitoneally administered a single dose of either polyethylene (3-75 μm in diameter) or polypropylene (4-50 μm in diameter) dusts at 50 mg/kg (10-15 mg/animal). No fibrosis were observed at 1 or 3 months after treatment in either treated group. On the other hand, chrysotile asbestos at lower doses produced characteristics of fibrotic nodules after 1 month and diffuse fibrosis by 3 months. It is difficult to evaluate these findings in view of the small numbers of animals used in this study and short duration of the study. Moreover, it was not clear whether the test dusts were fibrous or nonfibrous.

IV.3.2.2.2. Intratracheal Insufflation Studies

In the long-term study by MB Research Laboratories (1980) that was described in the oncogenicity section, lung fibrosis was not found in rats at 21 months following intratracheal insufflation of ozonized polyethylene SHFF, ozonized polypropylene SHFF, or HHF polypropylene. No information with regard to the characteristics of the test materials, the particle size distribution and administered doses was provided. These findings are therefore inadequate for definitive assessment.

IV.3.3. In Vitro Studies

IV.3.3.1. Genotoxicity

Polyethylene extracts were tested in the Salmonella/mammaliam activation assay in strains TA98, TA100 and TA1537 (Fevolden and Moller, 1978). This report is an abstract that provides no details, therefore it is not completely adequate for assessment.

IV.3.3.2. Cytotoxicity

Results of a single *in vitro* study showed that polypropylene and polyethylene dusts were significantly less cytotoxic to alveolar or peritoneal macrophages than was chrysotile asbestos. However, the authors did not specify whether the materials tested were fibrous or nonfibrous.

In the study by Styles and Wilson (1973), peritoneal and pulmonary macrophages (10^6 cells/mL) obtained from male and female Wistar rats were treated with either polyethylene or polypropylene dust at 500 µg/mL. The particle size of polyethylene and polypropylene dusts ranged between 3-75 µm and 4-50 µm in diameter, respectively. Negative control cultures were untreated while positive controls were treated similarly with chrysotile asbestos. Results of cell culture experiments indicated that cells treated with asbestos had the highest mortality (10-60 percent) while those tested with either polyethylene or polypropylene dust had the lowest mortality (less than 2 percent to 5 percent), as measured by the ratio of percentage of living to dead cells after 1 and 2 hours post-incubation.

IV.3.4. Assessment of Health Effects

Available data are inadequate for a conclusive assessment of potential carcinogenic and fibrogenic effects of polyolefin fibers, but they do suggest that polyolefin microfibers may have low fibrogenic potentials. Because polyolefin fibers or pulp are generally not respirable, inhalation of these fibrous materials would pose little or no health hazard to humans. On the other hand, a health hazard potential for polyolefin microfibers may exist since these may be respirable.

IV.3.4.1. Oncogenicity

Polyolefin fibers are not classifiable as to human carcinogenicity (Category D) on the basis of inadequate evidence of carcinogenicity in animal studies and no human data.

There are no available epidemiological or animal inhalation studies that examine the oncogenic potential of polyolefin fibers. The results of a limited intratracheal insufflation and an apparently well-conducted intraperitoneal injection oncogenicity study have not provided conclusive evidence of carcinogenicity for polyolefin fibers in animals. In the long-term intratracheal insufflation study, both polyethylene and polypropylene fibers did not induce tumor in rats (MB Research Laboratories, 1980). However, the lack of information on the nature, size distribution and dosage of the test materials precludes any definitive assessment of the oncogenicity of these fibers under the conditions of the study. In the long-term intraperitoneal injection study in rats, a low tumor incidence

was obtained with polypropylene microfibers (Pott et al., 1987b). These results, however, were only preliminary and a full evaluation cannot yet be made.

In contrast, the results of subcutaneous or intraperitoneal implantation studies showed that polyethylene/polypropylene disc, film, rod or fragments cause local sarcomas in mice and rats (IARC, 1979). However, because the test materials were not in fibrous form, these findings are not considered relevant to the assessment of the oncogenicity of fibrous polyolefin per se.

IV.3.4.2. Fibrogenicity

There is no information available on the fibrogenic effects of polyolefin fibers in humans and animals via inhalation exposure. Available animal studies by the injection/insufflation method and a single in vitro cytotoxicity study provide inconclusive data, and thus definitive assessment of the fibrogenic potential of polyolefin fibers cannot be made, although these studies seem to suggest a lower fibrogenic potential than that of asbestos.

In a long-term intratracheal study with polyethylene and polypropylene fibers, lung fibrosis was not observed in rats following 21 months (MB Research Laboratories, 1980). In a short-term study, peritoneal fibrosis was not found in rats treated with either polyethylene or polypropylene dusts, whereas under similar experimental conditions but at lower doses, chrysotile asbestos induced a low level of fibrosis after 3 months (Styles and Wilson, 1973). These in vitro results are

supported by the finding of a single in vitro study that both polypropylene and polypropylene dusts had significantly lower cytotoxicity in rat alveolar/peritoneal macrophages than did chrysotile asbestos (Styles and Wilson, 1973). However, the lack of information on the characteristics of the tested fibers makes it difficult to draw any definitive conclusions for this fiber category. On the other hand, preliminary results of an apparently well-conducted long-term intraperitoneal injection study in rats with long, thin polypropylene fibers showed a strong degree of adhesions of the abdominal organs. However, without histological data, it is not yet known whether or not fibrosis was also induced by polypropylene fibers (Pott et al., 1987b).

IV.3.5. Recommendations

A chronic inhalation study is recommended to further evaluate the oncogenic and fibrogenic potential of polyolefin microfibers. Because of a low health hazard associated with inhalation exposure to polyolefin fibers and pulps, additional animal tests do not appear necessary at this time.

V. Mechanisms of Fiber-Induced Diseases: Relationships between Fiber Properties and Pathogenicity

Epidemiological and experimental evidence accumulated thus far suggests that inhalation of fibrous dust other than asbestos might also be associated with malignant and nonmalignant pulmonary diseases in humans. However, the carcinogenicity and fibrogenicity of nonasbestos fibers appear to be variable. While

erionite seems to be at least as potent as asbestos, if not more so, fibrous glass and mineral wools are probably less hazardous than asbestos. On the other hand, available experimental data suggest that ceramic aluminum silicate glass, ultrafine aramid (Kevlar®) fibrils, and long-fibered attapulgite are potentially pathogenic, but conclusive assessment of the comparative oncogenic potential of these fibers and that of asbestos cannot yet be determined. As for wollastonite, short-fibered attapulgite, carbon fibers, and polyolefin fibers, these fibers appear to exhibit considerably lower pathogenic potential than asbestos. It should be stressed, however, that these assessments are by no means definitive because of incomplete data bases.

Although it seems that certain asbestiform fibers can cause asbestos-related diseases, there is some evidence suggesting that the pattern of diseases may vary with different fiber types. For example, available epidemiological evidence suggests that erionite exposure appears to be associated mainly with malignant mesothelioma of the pleura and peritoneum. This is consistent with animal evidence showing that inhalation exposure to erionite produces very high rates of mesothelioma in animals. The development of lung cancer, however, has not been demonstrated experimentally nor has it been conclusively established in human studies. On the other hand, for man-made mineral fibers, there is some epidemiological evidence for a possible association of lung cancer and occupational exposure to fibrous glass or mineral wool, but a risk of mesothelioma is not apparent.

An explanation of these differences may lie in the different intrinsic fiber properties which may control biological activity. It should be pointed out, however, that mechanisms by which mineral fibers, including asbestos, produce pathogenicity are not understood. Furthermore, little is known of the physicochemical properties that determine pathogenicity. However, it is still important to briefly discuss the relationships of fiber characteristics with possible mechanisms of fiber-induced diseases so that speculation about their importance can be focused in terms of research needs, and qualitative ranking of the hazard of fibers.

Clearly, fiber dimension is an important determinant for the development of any type of fiber-induced disease because it governs the entry and bioavailability of a fiber at target tissues. Fiber diameter is the most important factor in determining the respirability of the fiber. The thinner the fiber, the more respirable it is and the more easily it can penetrate into the lung. Fiber length and shape also affect the respirability and pulmonary deposition of the fiber but to a lesser degree. However, fiber length is more important in terms of fiber retention. Short fibers (<5 μm) are readily cleared by macrophage uptake while long fibers (>20 μm) which are not efficiently removed by phagocytosis may be retained long enough to cause diseases.

Fiber retention is probably also determined by the biological solubility of the fiber. In the lung, asbestos and nonasbestos fibers may undergo physicochemical alterations to

varying degrees, which could result in fragmentation and dissolution. Thus, it would appear that fibers which have low solubility, i.e., more durable fibers, are potentially more hazardous because of their long retention at target tissues. It has therefore been suggested that ceramic fibers are of considerable concern because they are relatively durable. On the other hand, the lower hazard potential of mineral wool and fibrous glass might be due to their high solubility. It should be noted that the solubility of a fiber is probably largely determined by fiber chemical characteristics.

With respect to the role of fiber properties in mediating biological effects and the development of diseases, fiber size also appears important for the pathogenesis of malignant mesothelioma. In a series of studies by Stanton and co-workers (Stanton et al., 1977, 1981) involving intrapleural implantation of various fibrous dusts of diversified chemical, crystallographic and morphological structures, a correlation was demonstrated between tumor incidence and the number of fibers present with lengths greater than 8 μm and diameters less than 0.25 μm. These studies provided the foundation for the "long, thin" hypothesis (also known as the Stanton's hypothesis) for the pathogenesis of mesothelioma. This hypothesis is supported by most of the experiments conducted to date by other investigators, showing that regardless of fiber characteristics, longer, thinner fibers are more carcinogenic than short, thick fibers by a variety of intracavitary injection methods. Furthermore, for a given fiber type, samples containing more long, thin fibers are

considerably more carcinogenic than those with mostly shorter thin fibers.

Bertrand and Pezerat (1980) statistically reanalyzed the data obtained from Stanton's studies and showed that carcinogenesis is a result of a continuous function of the aspect ratio (ratio of fiber length to fiber diameter), and the effects of fiber length and diameter cannot be separated. Most of the fibrous dusts which have been reported to be carcinogenic have high aspect ratios, and therefore, tend to support this hypothesis. However, this hypothesis does not explain the fact that short, thin attapulgite fibers from various sources with high aspect ratios are not carcinogenic in animals by either the intrapleural or intraperitoneal injection method. The only attapulgite samples which have been shown to be carcinogenic are those containing considerable amounts of long, thin fibers. This finding appears to argue against the Betrand and Pezerat hypothesis but does support the role of fiber length in fiber carcinogenesis.

The fiber size hypothesis, however, cannot explain the differential carcinogenic responses observed for various fiber types with similar fiber size distributions. A most notable example is that of erionite, which has comparable fiber size distribution as that of asbestos, yet is more potent in inducing mesothelioma in animals fiber per fiber than asbestos by either inhalation or injection methods. Thus, it would appear that other fiber properties such as chemical constitution and/or surface properties are also important in the development of

mesothelioma. However, the fact that synthetic nonfibrous erionite, which has identical chemical composition as naturally-occurring fibrous erionite, is not carcinogenic argues against the direct role of chemical constitution but rather supports the importance of fiber morphology in fiber carcinogenesis. It should also be noted that other nonfibrous particles do not generally cause mesothelioma.

Mesothelioma, which is not known to be associated with cigarette smoking, could be mediated by different mechanism(s) than those by lung cancer. It has been postulated that mineral fibers may behave as complete carcinogens in mesothelial cells and fibroblasts, the progenitors of mesotheliomas and pleural sarcomas (Mossman et al., 1983). This hypothesis is supported by the observation that erionite, which is the most potent mesothelioma-inducing fibrous agent, is highly genotoxic while asbestos and other fibers such as glass fibers are only weakly genotoxic. Based on the limited data base, the genotoxicity of fibers also appear to be influenced by fiber dimension. Thin fibers (e.g., fibrous glass) generally show some degree of clastogenicity and cell transformation, whereas coarse fibers have little or no activity. Fiber length also appears to affect not only the ability of fibers to be phagocytized but also the ability of intracellular fibers to induce cytogenetic damage and cell transformation.

There is also evidence to indicate that fiber size appears to be important in the induction of lung cancer. Davis et al. (1986, 1987) recently demonstrated that long, thin amosite and

chrysotile asbestos fibers are more potent than short, thin fibers in inducing lung tumors in rats via inhalation. It would appear that the different tumorigenic responses between short versus long fibers could be explained by their differences in the lung residence time and biological activity. Long fibers of a number of mineral fibers have been shown to be retained longer in the lung than short fibers and, moreover, long fibers are generally more cytotoxic and genotoxic than short fibers.

Emphasis has also been placed on the importance of surface properties of asbestos regarding the cocarcinogenic or promotional ability of asbestos in the development of lung cancer. In humans, a potentiating increase in lung cancer risk associated with asbestos exposure among cigarette smokers has been well documented. A synergistic effect has also been demonstrated experimentally in animals exposed by intratracheal instillation to a combination of asbestos and chemical carcinogens such as polycyclic hydrocarbons (PAHs) found in cigarette smoke. It has been hypothesized that asbestos fibers might serve as a physical carrier of chemical carcinogens, providing a means for cellular transport and uptake (Mossman and Craighead, 1979). It would appear that while the surface area would influence the quantity of chemical carcinogen adsorbing to the fiber, the parameters which actually determine the adsorption of chemical carcinogens onto the fiber, would most likely be the specific surface chemical characteristics of the fiber.

It should be noted that a synergistic effect in the induction of lung cancer has not yet been observed between

cigarette smoke and exposure to other asbestiform fibers (e.g., fibrous glass, mineral wool). Furthermore, unlike asbestos, erionite does not increase the genotoxicity of chemical carcinogens (e.g., benzo[a]pyrene), and extraction of erionite with organic solvent to remove potential organic contaminants does not reduce its in vitro genotoxicity (Brown et al., 1987). These findings suggest that asbestos fibers might have different adsorptive properties that are not shared by other fibers, which enable them to act as promotional agents in the development of lung cancer.

Fiber length also appears to be an important determinant in the development of lung fibrosis. Available experimental studies have shown that long, thin asbestos fibers are more fibrogenic than short, thin fibers in animals via inhalation or intratracheal instillation. It has been postulated that the sequence of cellular events leading to fibrosis probably involves first the interaction between the fiber and macrophage followed by a macrophage-fibroblast direct interaction and/or via effects on an intermediary cell type (NRC, 1984). If the theory is correct, i.e., at least with regard to the initial step, then the observation that long fibers are generally more cytotoxic to macrophages than are short fibers does indeed support the importance of fiber length in the development of fibrosis. However, the fact that long, thin fibers of various types, such as those of asbestos and fibrous glass, do not necessarily have comparable fibrogenic activity, and that respirable nonfibrous particles (e.g., silica) are also fibrogenic indicate that dust

particle characteristics other than morphology and size may also play an important role in the induction of fibrosis.

In conclusion, it is now recognized that the inhalation of durable fibers of certain diameter and length size range may be associated with the development of malignant and nonmalignant lung diseases. However, the pathogenic response may vary depending on the nature of the fibrous dust, including chemical constitution, solubility, surface charge, surface area, fiber size and morphology. Because these properties are probably interrelated, elucidation of the pathogenicity and mechanisms of fiber-induced diseases has been difficult. To increase our understanding of the health hazards of fibrous dusts, it is therefore necessary to study the common physical and chemical properties of these fibers in relation to their biological activity. This could lead to modifications of the physicochemical properties of fibers in order to minimize the production of adverse effects without affecting the desired properties necessary for industrial and commercial usage. Until the gaps in knowledge of this subject are filled, the fiber size model remains useful for further experimental studies, as well as environmental and epidemiological studies. Additional studies should also be conducted to elucidate the mechanisms for fiber-induced cytotoxicity, genotoxicity, and pathogenicity. Further studies investigating the relationship between fibrogenicity and carcinogenicity are also needed.

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VII. APPENDIX

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIBROUS GLASS

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Glass Wool	20% <2 μm diameter; average diameter near 5 μm; 70% >5 μm long	Guinea Pigs Rats	100 50	Inhalation	0.145 mg/m^3 0.03 mg/m^3	44 mo 28 mo	44 mo 28 mo	Little evidence of dust reaction in any species; No fibrosis; no pulmonary tumors	Poor animal survival; low concentration level	Schepers (1955), 1959a, 1959b, 1961) Schepers and Delahunt (1955); Schepers (1976)
Coated Glass Fibers (calcium carbonate)	Not specified	Monkeys Guinea Pigs Rabbits Rats	5 40 18 30	Inhalation	4.6 mg/m^3	8 mo 24 mo 18 mo 18 mo	8 mo 24 mo 18 mo 18 mo	As above	No information on dust cloud	As above
Coated Glass Fibers (calcium sulfate)	Not specified	Guinea Pigs Rabbits Rats	63 18 20	Inhalation	3.8 mg/m^3	15 mo 15 mo 9 mo	15 mo 15 mo 9 mo	As above	No information on dust cloud	As above
Glass Fibers (uncoated)	Average diameter 0.5 μm; average length 10 μm (5-20 μm)	Rats Hamsters	30 30	Inhalation	135 mg/m^3	2 yr	Lifespan	No fibrosis or tumors; histological changes limited to those seen from inert dusts	Poor survival	Gross et al. (1970) Gross (1976)
Coated Glass Fibers (Phenol formaldehyde resin)	As Above	Rats Hamsters	30 30	Inhalation	106 mg/m^3	2 yr	Lifespan	As above		As above
Coated Glass Fibers (starch binder)	As Above	Rats Hamsters	30 30	Inhalation	113 mg/m^3	2 yr	Lifespan	As above		As above

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIBROUS GLASS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Fibrous Glass	80% <1 μm diameter; 92% <10 μm long	Male Albino Mice (Charles River, CDR-1)	20	Inhalation	1070 fibers/mL	6 weeks	6 weeks	No lung damage; no fibrosis	Short exposure	Morisset et al. (1979)
Fiberglass insulation	80% were 2.5 μm diameter and 6-11 μm long	Male A-strain Mice	12	Mixed in bedding material (inhalation)	300 mg/ 100 mg bedding	30 days	90 days	Bronchogenic tumor (3/12); septal cell tumor (2/12)	No unexposed controls; short exposure; small number of exposed animals; atypical method of administration	Morrison et al. (1981)
Fiberglass	Average diameter 1.2 μm; length <2 μm; 7% with aspect ratio >3	Male Charles River Sprague-Dawley Rats	46	Inhalation	400 mg/m^3 (700 fibers/mL)	90 days	18-24 mo	Bronchoalveolar adenoma (2/19); control rats had no tumors (0/13); No fibrosis	Short exposure; small number of animals; mostly nonfibrous material	Lee et al. (1979) Lee et al. (1981)
		Male Albino Guinea Pigs	32	Inhalation	400 mg/m^3 (700 fibers/mL)	90 days	18-24 mo	Bronchoalveolar adenoma (2/8); control animals had no tumors (0/6); no fibrosis		As above
Glass Fibers (Johns-Manville Code 100) Uncoated Glass Wool Coated Glass Wool	Not specified	SPF Fischer Rats	2/group	Inhalation	10 mg/m^3	50 weeks	16 mo	Focal fibrosis was evident with all the dusts but more marked with chrysotile; noncoated glass wool more reactive than uncoated glass wool	Small number of animals	Johnson and Wagner (1980)

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIBROUS GLASS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Glass Microfibers (A blend of JM C102 and JM C104)	Median diameter 0.6 μm; median length 6.25 um	Male Baboons (Papio ursinus)	Not specified (10 animals in total including positive controls)	Inhalation	7.54 mg/m^3 (1122 fiber/mL)	35 mo	41-42 mo	Fibrosis after 18 and 30 mo exposure; however, lesions less severe than those in animals exposed to crocidolite; no evidence of malignancy	Short exposure in relation to lifespan of baboons; no unexposed controls	Goldstein et al. (1983) Goldstein et al. (1984)
Glass Microfibers (Johns-Manville Code 100)	Mean diameter 0.3 μm; 71% <10 μm long	Male and Female Fischer 344 Rats	28 of each sex	Inhalation	10 mg/m^3 (1436 fibers/mL)	12 mo	Lifespan	No fibrosis at 24 mo; lung adenocarcinoma (1/48)	No tumors in unexposed controls; 12 tumors in 48 chrysotile exposed animals (11 lung adenocarcinomas; 1 adenoma)	Wagner et al. (1984)
Glass Wool (uncoated)	0.2-3 μm diameter; 70% >10 μm long	Male and Female Fischer 344 Rats	28 of each sex	Inhalation	10 mg/m^3 (240 fibers/mL)	12 mo	Lifespan	No fibrosis; lung adenocarcinoma (1/48)		As above
Glass Wool (coated)	0.2-3 μm diameter; 57% >10 μm long	Male and Female Fischer 344 Rats	28 of each sex	Inhalation	10 mg/m^3 (323 fibers/mL)	12 mo	Lifespan	No fibrosis; lung adenoma (1/48)		As above
Glass Microfibers (JM Code 100)	Not available but presumably similar to that used in Wagner et al. (1984)	Male and Female Fischer 344 Rats	Unspecified	Inhalation	10 mg/m^3	12 mo	Lifespan	No fibrosis; no neoplasms (0/55); lung adenocarcinoma in 2 of 53 negative controls	Lung tumors in 11 of 56 chrysotile exposed rats (7 adenocarcinoma; 4 adenoma)	McConnell et al. (1984)

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIBROUS GLASS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Glass Microfibers (JM Code 100)	51% with 0.2-0.5 μm diameter	Male and Female IOPS AF/Han Rats	24 of each sex	Inhalation	5 mg/m^3 (332 fibers/mL)	12 or 24 mo	24 mo	No fibrosis; no neoplasms (0/48)	Low airborne fiber concentration compared to chrysotile; chrysotile exposed animals with lung tumors in 9/47 (histology not provided)	Le Bouffant et al. (1984)
Glass Wool (uncoated)	68% <1 μm diameter	Male and Female IOPS AF/HAN Rats	24 of each sex	Inhalation	5 mg/m^3 (48 fibers/mL)	12 or 24 mo	24 mo	No fibrosis; pulmonary tumors in 1 of 45 exposed rats		
Glass Microfibers	0.45 μm mean diameter	Male Syrian Hamsters	69	Inhalation	3 mg/m^3 (3,000 fibers/mL)	24 mo	Lifespan	No fibrosis or lung neoplasms	Bronchoalveolar tumor in 1 sham hamster (1/58); bronchometaplasia in crocidolite asbestos exposed hamsters; three cases of neoplasms in crocidolite exposed rats (1 mesothelioma, 2 bronchoalveolar tumors); no tumors in sham control or unexposed rats	Smith et al. (1984) Smith et al. (1986)
			70		0.3 mg/m^3 (300 fibers/mL)					
		Female Osborne-Mendel Rats	57	Inhalation	3 mg/m^3 (3,000 fibers/mL)	24 mo	Lifespan	No fibrosis or lung neoplasms		
			57		0.3 mg/m^3 (300 fibers/mL)					

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIBROUS GLASS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Glass Wool	3.1 μm mean diameter	Male Syrian Hamsters	60	Inhalation	10 mg/m^3 (100 fibers/mL)	24 mo	Lifespan	No fibrosis or lung neoplasms in exposed rats or hamsters	Glass wool aerosol dusts had a large proportion of nonfibrous material	Smith et al. (1986)
		Female Osborne-Mendel Rats	52							
Glass Wool	5.4 μm mean diameter	Male Syrian Hamsters	66/65	Inhalation	12 mg/m^3 (100 fibers/mL) or 1.2 mg/m^3 (10 fibers/mL)	24 mo	Lifespan	No fibrosis or lung neoplasms in exposed rats or hamsters		As above
		Female Osborne-Mendel Rats	57/61							
Glass Wool	6.1 μm mean diameter	Male Syrian Hamsters	99	Inhalation	9 mg/m^3 (25 fibers/mL)	24 mo	Lifespan	No fibrosis or lung neoplasms in exposed rats or hamsters		As above
		Female Osborne-Mendel Rats	58							

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIB[redacted]ASS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Fibrous Glass (Red Binder; Group I)	4.6 μm diameter; >20 μm long	Male and Female Fischer 344 Rats	50/sex	Inhalation	15 mg/m^3	86 weeks	86 weeks	No fibrosis; no pulmonary tumors or mesothelioma in any treated rat or monkey groups; statistical increase in mononuclear cell leukemia in Group I female rats (p=0.047; Fisher exact test), Group III (p=0.024) and Group IV (p=0.002) male rats.	Early death in 37% rats in treated and control group; short treatment and study duration	Mitchell et al. (1986)
		Male Cynomolgus Monkeys	15	Inhalation	15 mg/m^3	72 weeks	72 weeks			
Fibrous Glass (Yellow Binder; (Group II)	0.5-3.5 μm diameter; >10 μm long	Male and Female Fischer 344 Rats	50/sex	Inhalation	15 mg/m^3	86 weeks	86 weeks			As above
		Male Cynomolgus Monkeys	15	Inhalation	15 mg/m^3	72 weeks	72 weeks			
Fibrous Glass (Uncoated; Group III)	<3.5 μm diameter; >10 μm long	Male and Female Fischer 344 Rats	50/sex	Inhalation	5 mg/m^3	86 weeks	86 weeks			As above
		Male Cynomolgus Monkeys	15	Inhalation	5 mg/m^3	72 weeks	72 weeks			
Fibrous Glass (Uncoated; Group IV)	<3.5 μm diameter; <10 μm long	Male and Female Fischer 344 Rats	50/sex	Inhalation	5 mg/m^3	86 weeks	86 weeks			As above
		Male Cynomolgus Monkeys	15	Inhalation	5 mg/m^3	72 weeks	72 weeks			

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIBR[redacted]ASS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Ibrous Glass	6 samples of varying fiber size distributions	Female pathogen-free Osborne-Mendel Rats	Not specified	Intrapleural implantation	40 mg	Single dose	2 yrs	Moderately high incidence of mesothelioma in two samples of fine fiberglass milled to approach length of asbestos fibers	High incidence of mesothelioma in amosite, chrysotile, and crocidolite exposed group	Stanton and Wrench (1972)
Iberglass Code 110)	30% <2.5 µm diameter; 60% >20 µm long	SPF Wistar Rats (sex unspecified)	36	Intrapleural injection	20 mg	Single dose	Lifespan	No mesothelioma	Mesothelioma in 23/36 rats treated with SFA chrysotile	Wagner et al. (1973)
Glass Fibers	3.5 µm diameter; <20 µm long 3.5 µm diameter; >100 µm long 0.05 µm diameter; <20 µm long 0.05 µm diameter; >100 µm long	Balb/C Mice	25/group	Intrapleural injection	10 mg	Single dose	18 mo	Long fiber samples produced massive fibrosis while short fibered samples produced only discrete granulomas with minimal fibrosis		Davis (1976)
Ibrous Glass	17 samples of diverse dimensional distributions	Female Osborne-Mendel Rats	30 in each treated group	Intrapleural implantation	40 mg	Single dose	2 yrs	Highest yield of pleural sarcoma with fibers <1.5 um in diameter and >8 um in length		Stanton et al. (1977)

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIBR[redacted]SS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Glass Fiber (Code 110)	17% <1 μm diameter; median diameter 1.8 μm; median length 22 μm	Wistar Rats (Male and Female)	Total of 32	Intrapleural injection	20 mg	Single dose	Lifespan	No mesothelioma	No tumors in saline treated controls	Wagner et al. (1976)
Glass Microfiber (Code 100)	99% <0.5 μm diameter; median diameter 0.12 μm; median length 1.7 μm; 2% >20 μm long	Wistar Rats (Male and Female)	Total of 32	Intrapleural injection	20 mg	Single dose	Lifespan	Pleural tumors in 4 of 32 rats (p=0.01)		As above
Fibrous Glass	Mean diameter 0.1 μm; 82% >20 μm long	Male Syrian Golden Hamsters	60	Intrapleural injections	25 mg	Single dose	Not specified	Intrathoracic tumors (9/60)	Histology not provided; no control group reported	Smith et al. (1980)
	Median diameter 0.09 μm; 0-2% >20 μm long							No tumors		
	Mean diameter 0.33 μm; 46% >20 μm long							Intrathoracic tumors (2/60)		
	Mean diameter 0.41 μm; 0-2% >20 μm long							No tumors		
	Mean diameter 1.23 μm; 34% >20 μm long							Intrathoracic tumors (2/60)		
	Mean diameter 1.49 μm; 0-2% >20 μm long							No tumors		

TABLE I. SUMMARY OF ANIMAL STUDIES ON FI[redacted]SS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Fine Glass Fibers	Mean diameter 0.229 μm; mean length 5.8 μm	Male Sprague-Dawley Rats	45	Intrapleural injection	20 mg	Single dose	Lifespan	13% incidence of pleural tumors (6/44)	No tumors in vehicle controls (saline); high incidence of mesothelioma in chrysotile (45%) and crocidolite (54%) treated animals	Lafuma et al. (1980); Monchaux et al. (1981)
Fibrous Glass	22 samples of diverse dimensional distributions	Female Osborne-Mendel Rats	30-50	Intrapleural implantation	40 mg	Single dose	2 years	Probability of pleural sarcoma best correlated with the number of fibers with diameters <0.25 um and length >8 um; relatively high correlations also noted for fibers with diameters up to 1.5 um and length >4 um		Stanton et al. (1981)
Glass Microfibers (Johns-Manville Code 100)	86% <0.6 μm diameter; 88% <5 μm long	SPF Sprague-Dawley Rats	48	Intrapleural injection	20 mg	Single dose	Lifespan	Pleural mesothelioma in 4 of 48 rats	Six cases of pleural tumors in 48 rats treated with UICC chrysotile asbestos	Wagner et al. (1984)
Glass Wool (uncoated)	85% <1 μm diameter; 76% <10 μm long	SPF Sprague-Dawley Rats	48	Intrapleural injection	20 mg	Single dose	Lifespan	No pleural tumors		As above
Glass Wool (with resin)	85% <1 μm diameter 77% <10 μm long	SPF Sprague-Dawley Rats	48	Intrapleural injection	20 mg	Single dose	Lifespan	Pleural tumor in 1 of 48 rats		As above

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIBROUS GLASS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Fibrous Glass (S + S106)	Mean diameter 0.5 µm; 72.6% <5 µm long	Female Wistar Rats	40	Intraperitoneal injection	2 mg	Single dose	Not specified	Abdominal tumors (mostly mesothelioma) 2.5% at 2 mg, 10% at 10 mg and 57% at 100 mg; fibrosis in animals receiving 100 mg of glass fibers; less severe lesions at 10 mg and 2 mg dose levels	Tumor rates of 15-67% in UICC chysotile asbestos treated group at similar dosing regimen; latency period inversely related to dose; no tumors in saline treated controls	Pott and Friedrichs (1972); Pott et al. (1974) and (1976)
					10 mg	Single dose				
					4x25 mg	Four doses at weekly intervals				
Fiberglass (MN104)	50% <0.2 µm diameter; 50% <11 µm long	Wistar Rats (sex unspecified)	80	Intraperitoneal injection	2mg	Single dose	Not specified	Abdominal tumors (7 mesothelioma, 1 spindle cell sarcoma)		Pott et al. (1976)
					10 mg	Single dose		23 mesothelioma, 3 spindle cell sarcoma, 1 carcinoma		
					2 x 25 mg	Two doses at weekly intervals		47 mesothelioma, 6 spindle cell sarcoma, 2 polymorphocellular sarcoma		
Fiberglass (MN 112)	50% <1 µm diameter 50% <28 µm long	Wistar Rats (sex not specified)	40	Intraperitoneal injection	20 mg	Single dose		Peritoneal mesothelioma (4/40)		As above

TABLE 1. SUMMARY OF ANIMAL STUDIES ON FIBR[redacted]ASS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Glass Microfibers	Average diameter of 0.05 μm	Balb/c mice	25	Intraperitoneal injection	25 mg	Single dose	Not Specified	Peritoneal tumors (3/25)		Davis (1976)
		Rats (unspecified strain)	18	Intraperitoneal injection	10 mg	Single dose	Not Specified	Peritoneal tumors (3/28)		
Glass Fibers (JM 104)	Not specified	Wistar Rats (Ivanovas)	50	Intraperitoneal injection	10 mg	Single dose	Not specified	Abdominal tumors (25/49; 51%)	No experimental details	Pott et al. (1980)
		SIV Rats (Ivanovas)	50					Abdominal tumors (36/50; 72%)		
		Sprague-Dawley Rats (Hagemann)	50					Abdominal tumors (29/49; 59.2%)		
		Wistar Rats (Hagemann)	50					Abdominal tumors (39/49; 79.6%)		
Glass Microfibers (JM 104)	50% <0.3 μm diameter; 90% <13 μm long	Wistar/Sprague-Dawley Rats	40-60 group	Intraperitoneal injection	2 or 10 mg	Single dose	Not specified	Peritoneal sarcoma and mesothelioma (40-70%)		Pott et al. (1984)
Glass Microfibers (JM 100)	50% <0.3 μm diameter; 90% <7 μm long	Wistar/Sprague-Dawley Rats	40-60 group	Intraperitoneal injection	2, 5, or 10 mg	Single dose	Not specified	Peritoneal sarcoma and mesothelioma (2-10%)		As above
Fibrous Glass	0.45 μm mean diameter	Female Osborne-Mendel Rats	25	Intraperitoneal injection	25 mg	Single dose	Not specified	Abdominal mesothelioma (8/25); Abdominal reactive tissue/ fibrosis in 13/17 animals	Tumor incidence of 80% (20/25); in positive controls treated with UICC crocidolite asbestos; no tumors in saline controls or untreated animals	Smith et al. (1986)

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIBR[redacted]SS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Fibrous Glass (Uncoated)	Average diameter 0.5 μm; average length 10 μm	Rats (strain unspecified)	15/ 30	Intratracheal instillation	3 x 3.5 mg or 10 x 3.5 mg	Not specified	Lifespan	No tumors in any treated rat or hamster group	Experimental details not available; no positive controls; small number of animals	Gross (1976)
		Hamsters (strain unspecified)	12		3 x 3.5 mg					
Fibrous Glass (Coated with resin)	Average diameter 0.5 μm; average length 10 μm	Rats	30	Intratracheal instillation	3 x 3.5 mg or 10 x 3.5 mg	As above	As above		As above	As above
		Hamsters	12		1 x 3.5 mg					
			12		2 x 1.75 mg					
			12		3 x 3.5 mg					
Fibrous Glass (Coated with Starch Binder)	Average diameter 0.5 μm; average length 10 μm	Rats	15	Intratracheal instillation	3 x 3.5 mg					As above
			30		10 x 3.5 mg					
		Hamsters	12		3 x 3.5 mg					

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIBR[redacted]SS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Glass Fibers (Thin Fibers)	Diameter <1 μm; 93% <10 μm long	Guinea Pigs	30	Intratracheal instillation	2 x 12.5 mg	Biweekly	2 years	No fibrosis nor tumors	Long fibers (>10 um) of crocidolite asbestos produced fibrosis; short fibers (<10 um) did not produce fibrosis	Wright and Kuschner (1976, 1977)
	Diameter <1 μm; 92% >10 μm long	Guinea Pigs	30		3 x 4 mg	Biweekly	2 years	Fibrosis; no tumors		
Glass Fibers (Very Thin)	Diameter <0.3 μm and <5 μm long	Guinea Pigs	30	Intratracheal instillation	2 x 12.5 mg	Biweekly	2 years	No fibrosis nor tumors		As above
	Diameter <0.3 μm; 50% >10 μm long	Guinea Pigs	30		2 x 6 mg	Biweekly	2 years	Fibrosis; no tumors		
Glass Fibers (Thick)	Diameter 2 μm; 88% <10 μm long	Guinea Pigs	30	Intratracheal instillation	2 x 12.5 mg	Biweekly	2 years	No fibrosis nor neoplasms		As above
	Diameter 2 μm 75% >10 μm long	Guinea Pigs	30		2 x 12.5 mg	Biweekly	2 years	No fibrosis nor neoplasms		

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIBR[redacted]SS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Uncoated Glass Microfibers	0.1 μm mean diameter	Male Syrian Hamsters	20	Intratracheal instillation	A total of 7 mg	Biweekly	11 months	Mild fibrosis in glass microfibers treated group at 11 months after instillation		Pickrell et al. (1983)
	0.25 μm mean diameter		20		2 mg	Weekly				
Household Insulation Glass fibers	2.3 μm mean diameter		20		21 mg	Not specified		No fibrosis		As above
	3.0 μm mean diameter		20		18 mg	Not specified		No fibrosis		
	4.1 μm mean diameter		20		17 mg	Not specified		No fibrosis		
Glass Microfibers (JM Code 104)	50% <0.3 μm diameter; 50% <7 μm long	Male Syrian Golden Hamsters	136	Intratracheal instillation	8 x 1 mg	Weekly	136 weeks	Lung carcinoma (5/136) Pleural mesothelioma (37/136) Thoracic sarcomas (6/136)	Tumors in crocidolite asbestos positive controls Lung carcinoma (9/142) Mesothelioma (8/142) Thoracic sarcoma (1/142)	Mohr et al. (1984)
	50% <0.3 μm diameter; 50% <4.2 μm long		138	Intratracheal instillation	8 x 1 mg	Weekly	136 weeks	Lung carcinoma (6/138) Mesothelioma (26/138) Thoracic sarcomas (6/138)	2 cases of thoracic sarcoma (2/135) in titanium dioxide treated group but no mesothelioma or lung neoplasms; no saline control group	

TABLE I. SUMMARY OF ANIMAL STUDIES ON FIBR[redacted]SS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Glass Microfibers (JM Code 104)	31% <0.25 μm diameter; 58% <5 μm long	Male and Female Syrian Golden Hamsters	35/sex	Intratracheal Instillation	26 x 1 mg	Biweekly for 52 weeks	85 weeks	No tumors (0/34 males; 0/30 females)	No positive control animals	Feron et al. (1985)
Fibrous Glass	0.45 μm mean diameter	Female Osborne-Mendel Rats	32	Intratracheal Instillation	5 x 2 mg	Weekly	Lifespan	Signficant fibrosis (7/22); no tumors	No lesions in saline controls and untreated animals	Smith et al. (1986)
Glass Microfibers (JM 104/ Sempstan)	50% <3.2 μm length; 50% <0.12 μm diameter	Female Wistar Rats	34	Intratracheal 0.5 mg Instillation	20 x 0.05 mg	Weekly	Lifespan	Lung tumors in 5/34 animals (1 adenoma, 2 adenocarcinoma, 2 squamous cell carcinomas) and 9/142 lung cancer	Positive control animals treated with crocidolite had 11/35 lung tumor	Pott et al. (1987a)

Table 2. SUMMARY OF ANIMAL [redacted] ON MINERAL WOOL

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Rock wool	58% <1 μm diameter 64% >10 μm long	Male and Female F344 rats	56	Inhalation	10 mg/m^3 (227 fibers/mL; for d <3 μm; and l >5 μm)	12 mo	Lifespan	No lung fibrosis; lung adenoma (2/48)	No tumors in unexposed controls; lung adenocarcinoma (11/48) and adenoma (1/48) in chrysotile asbestos positive controls	Wagner et al. (1984)
Saint-Gobain rock wool	22.7% <1 μm diameter; 60% >10 μm long	Male and Female Wistar IOPS rats	48	Inhalation	5 mg/m^3 (11 fibers/mL; for l >5 μm)	24 mo	24 mo	No pulmonary changes; no lung tumors (0/24 males; 0/23 females)	Very low fiber concentration; dust cloud contained mostly nonfibrous particles; nine cases of lung tumors in Canadian chrysotile asbestos control group (males, 5/24; females, 4/23); no tumors in unexposed control animal (0/27)	Le Bouffant et al. (1984)

Table 2. SUMMARY OF ANIMAL STUDIES [redacted]ERAL WOOL (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Mineral Wool	mean diameter 2.7 μm; 75% >10 μm long	Female Osborne-Mendel rats	55	Inhalation	12 mg/m^3 (200 fibers/mL; 76 fibers/mL for fibers longer than 10 μm with diameters <1 μm)	24 mo	Lifespan	No pulmonary fibrosis; No lung tumors (0/55)	3/57 UICC crocodolite positive control rats developed tumors (1 mesothelioma, 2 bronchoalveolar tumors); No tumors in sham controls (0/59) and unexposed rats (0/125)	Smith et al. (1986)
		Male Syrian hamsters	69	Inhalation	as above	24 mo	Lifespan	No lung fibrosis; no lung tumors (0/69)	No tumors in UICC crocodolite asbestos exposed group (0/58) or in unexposed controls (0/112); one bronchoalveolar tumor (1/58) in sham control group	

Table 2. SUMMARY OF ANIMAL STUDIES OF MINERAL WOOL (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Rock Wool (with resin)	77% <1 μm in diameter; 70% <5 μm long	SPF Fischer 344 rats (sex unspecified)	48	Intrapleural injection	20 mg	Single dose	Lifespan	Mesotheliomas (3/48)	Mineral wool samples contained more non-fibrous particles than fibrous material; UICC chrysotile asbestos produced 6 cases (6/48) of mesothelioma.	Wagner et al. (1984)
Rock wool (without resin)	82% <1 μm in diameter; 69% <5 μm long	SPF Fischer 344 rats (sex specified)	48	Intrapleural injection	20 mg	Single dose	Lifespan	Mesothelioma (2/48)		As above
Slag Wool (with resin)	70% <1 μm in diameter; 67% <5 μm long	SPF Fischer 344 rats (sex unspecified)	48	Intrapleural injection	20 mg	Single dose	Lifespan	No mesothelioma		As above
Slag Wool (without resin)	82% <1 μm in diameter; 80% <5 μm long	SPF Fischer 344 rats (sex unspecified)	48	Intrapleural injection	20 mg	Single dose	Lifespan	No mesothelioma		As above

Table 2. SUMMARY OF ANIMAL STUDIES [redacted]INERAL WOOL (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Rock Wool	50% <0.64 μm in diameter; 50% <4.1 μm long	Female Sprague-Dawley rats	45	Intraperitoneal injection	10 mg	Single dose	15 mo	No peritoneal tumors	Short observation period (ongoing study)	Pott et al. (1984)
Rock Wool	50% <1.90 μm diameter; 50% <23 μm long	Female Sprague-Dawley rats	63	Intraperitoneal injection	25 mg	3 x 25 mg	Lifespan	Peritoneal sarcoma/ mesothelioma (16%)	No positive control groups	As above
Slag Wool	90% <0.28 μm diameter; 9% <10 μm long	Female Wistar rats	41	Intraperitoneal injection	5 mg	Single dose	Lifespan	Peritoneal tumors (5%)	Small dose; tumor yield not statistically significant	As above
Basalt Wool	50% <0.52 μm diameter; 50% <58 μm long	Female Wistar rats	45	Intraperitoneal injection	5 mg	Single dose	15 mo	No peritoneal tumors	Relatively small dose; short observation period	As above
Basalt Wool	50% <1.8 μm diameter, 50% <20 μm long	Female Wistar rats	53	Intraperitoneal injection	15 mg	5 weekly doses	Lifespan	Peritoneal tumors in 30/53; negative saline control had a tumor incidence of 1/102; UICC/ Canadian chrysotile produced high incidence of tumor at much lower doses	Relatively large fibers; very high dose level; preliminary results only	Pott et al. (1987b)

Table 3. SUMMARY OF ANIMAL [redacted]ON CERAMIC FIBERS

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Ceramic Aluminum Silicate Glass	90% <3 μm long and <0.3 μm diameter	SPF Wistar rats (AF/HAN strain; sex unspecified	48	Inhalation	8.4 mg/m^3 (95 fibers/mL; d <3 μm, l >5 μm)	12 mo	Up to 32 mo	Interstitial fibrosis occurred to a lower but not significantly different degree than that for chrysotile asbestos animals observed in other studies; lung tumors in 8 animals (1 adenoma, 3 carcinomas, 4 malignant histiocytomas); no tumors in 40 unexposed control animals	Dust cloud contained mostly short, thin fibers	Davis et al. (1984)

Table 3. SUMMARY OF ANIMAL STUDIES [redacted]MIC FIBERS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Refractory Ceramic Fiber	83% > 10 µm long and 86% <2 µm diameter	Female Osborne-Mendel rats	55	Inhalation	12 mg/m^3 (200 fibers/mL)	24 mo	Lifespan	No lung fibrosis; no lung tumors; no tumors in sham or unexposed control	Low tumorigenic response in UICC crocidolite asbestos control rats (3/59; 1 mesothelioma, 2 bronchoalveolar tumor)	Smith et al. (1986)
		Male Syrian hamsters	70	Inhalation	12 mg/m^3 (200 fibers/mL)	24 mo	Lifespan	No lung fibrosis; one mesothelioma; no primary lung tumors; no tumors in unexposed control; one case of bronchoalveolar tumor in sham control	No tumors (0/58) in positive control hamsters exposed to UICC crocidolite asbestos	

Table 3. SUMMARY OF ANIMAL STUDIES [redacted]AMIC FIBERS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Refractory Alumina Fibers (as manufactured or "thermally aged")	Median diameter of 3.0 μm; median length 10.5-62 μm	Male and female albino rats of the Alderly Park (Wistar derived strain)	25 of each sex/group	Inhalation	2.18-2.45 mg/m^3	86 weeks	>86 weeks	Pulmonary reaction to both forms of Saffil was minimal; no pulmonary neoplasms	Levels of respirable dust in the atmospheres were low; positive control animals exposed to UICC chrysotile asbestos developed pulmonary neoplasms (9/38)	Pigott et al. (1981)
Synthetic Aluminum Silicate Fibers	Diameter between 0.5 and 1.0 μm; length unspecified	SPF Wistar rats	24 males 12 females	Intrapleural inoculation	20 mg	Single dose	Lifetime	Mesothelioma in 3/31 rats	Carcinogenic potency of ceramic fibers were considerably less than SFA chrysotile asbestos	Wagner et al. (1973)

Table 3. SUMMARY OF ANIMAL STUDIES [redacted]RAMIC FIBERS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Alumina Oxide Fibers (glass 21)	Not specified	Female Osborne-Mendel rats	50	Intrapleural implantation	40 mg	Single dose	2 yr	Pleural neoplasms in 2/47 rats	Tumor incidence not statistically significant	Stanton et al. (1981)
Zirconia oxide fibers (glass 22)	Not specified	Female Osborne-Mendel rats	50	Intrapleural implantation	40 mg	Single dose	2 yr	Pleural neoplasm in 1/45	Tumor incidence not statistically significant	
Refractory Alumina Fiber (Saffil)	Type A: median diameter 2.75 μm; median length 15.5 μm Type B: median diameter 3.7 μm; median length 17 μm	SPF Wistar rats (Alderley Park Strain)	22 males and 12 females per group	Intraperitoneal injection	20 mg	Single dose	up to 12 mo	Mild chronic inflammatory response with a mild amount of collagen in the abdominal tissues	Progressive peritoneal fibrosis in rats receiving UICC Rhodesian chrysotile asbestos	Pigott and Ishmael (1981)

Table 3. SUMMARY OF ANIMAL STUDIES [redacted]ERAMIC FIBERS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Saffil Alumina Fibers	median diameter 3.6 μm; median length 17 μm	SPF albino Wistar rats (Alderley Park Strain)	40 (20 of each sex)	Intraperitoneal injection	20 mg (0.2 mL of a 10% suspension of fibers)	Single dose	6 mo	Nodular deposit of connective tissue; no fibrosis	Marked peritoneal fibrosis in rats treated with UICC chrysotile asbestos	Styles and Wilson (1976)
Saffil Zirconia Fibers	median diameter 2.5 μm; median length 11 μm	SPF albino Wistar rats (Alderley Park Stain)	40 (20 of each sex)	Intraperitoneal injection	20 mg	Single dose	6 mo	Nodular deposit of connective tissue containing collagen		As above
Ceramic Aluminum Silicate Glass	90% <3 μm long and <0.3 μm diameter	SPF Wistar rats of AF/HAN stain (sex unspecified)	32	Intraperitoneal injection	25 mg	Single dose	Lifetime	Peritoneal neoplasms in 3/32 animals; first tumor occurred 850 days after injection	No vehicle control group	Davis et al. (1984)
Ceramic "Fiberfrax"	50% <8.3 μm long; 50% <0.91 μm in diameter	Female Wistar rats	47	Intraperitoneal injection	9 mg	5 weekly doses	Lifespan	Abdominal tumors in 33/47 animals; total number of fibers injected (173 x 10^6) were comparable to that of chrysotile at 0.25 mg (202 x 10^6)	Preliminary results; UICC chrysotile induced dose-related increase in peritoneal tumors at much lower doses (11/36, 21/34, 30/36 at 0.05, 0.25; and 1.0 mg, respectively	Pott et al. (1987b)

Table 3. SUMMARY OF ANIMAL STUDIES OF CERAMIC FIBERS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Ceramic MAN	50% < 6.9 μm in length; 50% <1.1 μm in diameter	Female Wistar rats	54	Intraperitoneal injection	15 mg	5 weekly doses	Lifetime	Abdominal tumors in 11/54 animals (Total number of fibers injected were 105×10^6)	UICC chrysotile asbestos induced dose-related increase in tumors at much lower doses	Pott et al. (1987b)
Refractory ceramic fibers	mean diameter 1.8 μm	Male Syrian hamsters	25 per group	Intraperitoneal injection	25 mg	Single dose	Lifetime	Abdominal mesotheliomas (2/25 and 5/21)	UICC crocidolite asbestos produced abdominal mesothelioma in 8/25 hamsters and 20/25 rats	Smith et al. (1986)
		Female Osborne-Mendel rats	25	Intraperitoneal injection	25 mg	Single dose	Lifetime	Abdominal mesotheliomas in 19/25 animals		
Refractory ceramic fibers	mean diameter 1.8 μm	Male Syrian hamsters	25	Intratracheal instillation	2 mg	weekly for 5 weeks	Lifetime	No primary lung tumors (0/25)	UICC crocidolite asbestos produced primary lung tumors in 20/27 hamsters and 2/25 rats; significant lung fibrosis	Smith et al. (1986)
		Female Osborne-Mendel rats	25	Intratracheal instillation	2 mg	Weekly for 5 weeks	Lifetime	Bronchoalveolar metaplasia in 6/22 rats; no lung tumors (0/22)		
Ceramic Aluminum Silicate Glass	mean diameter 2 μm; 50%< 75 μm long	Guinea pig (sex, strain unspecified)	Not specified	Intratracheal injection	25 mg	Single dose	6 weeks	Formation of ferruginous bodies and large granulomas; no fibrosis	Short study	Davis et al. (1970)

Table 4. SUMMARY OF ANIM[redacted]IES ON ERIONITE

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Oregon Erionite	86% <0.4 μm diameter; 92% <10 μm long	Male and Female Fischer 344 rats	20 of each sex	Inhalation	10 mg/m^3 (354 fibers/mL)	12 months	Lifespan	Pleural mesothelioma in 27/28 animals; considerable shortening of latency period (mean survival time of 580 days)	No mesothelioma and only 1 lung tumor in 28 crocidolite-exposed animals; synthetic nonfibrous zeolite produced 1/28 mesothelioma and 1/28 lung tumor	Wagner et al. (1985)
Oregon Erionite	75% <6 μm long; 92% <0.2 μm diameter	Fischer 344 rats	20 of each sex	Intrapleural injection	20 mg	Single dose	Lifespan	40 pleural mesothelioma (100%); mean survival time of 390 days	Chrysotile produced 19/40 mesothelioma with a mean survival time of 678 days; vehicle control animals had one case of mesothelioma	Wagner et al. (1985)
Karain Rock fiber	91% <0.2 μm diameter; 86% <6 μm long	Fischer 344 rats	20 of each sex	Intrapleural injection	20 mg	Single dose	Lifespan	38 pleural mesothelioma (95%); mean survival time 434 days		As above
Erionite	Not specified	Sprague-Dawley rats	20 of each sex	Intrapleural injection	25 mg	Single dose	53 weeks	Mesothelioma in 9/40 animals after 53 weeks; fibrogenic reactions	Interim results only; no pleural tumors in crocidolite asbestos group and vehicle controls	Maltoni et al. (1982a)

Table 4. SUMMARY OF ANIMAL STUDIES OF ERIONITE (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Erionite	20% <1 μm and 95% <8 μm long; 19% <0.1 μm and 95% <1 μm diameter	Male Swiss albino mice	12	Intraperitoneal injection	10 mg	Single dose	Six animals sacrificed at 2-3 months; remaining 6 animals were maintained until death (up to 15 months	Malignant peritoneal tumors in 2/5 animals after 15 months; fibrotic lesions presented at neoplastic tissues		Suzuki (1982)
		Male Swiss albino mice	5/group	Intraperitoneal injection	10 mg or 30 mg	Single dose	Not Specified	Significant fibrosis; malignant peritoneal tumors in 4/5 animals receiving 10 mg of erionite; animals receiving 30 mg erionite died with intestinal obstruction	Chrysotile (10 mg) produced peritoneal tumors in 2/5 animals; no tumors in untreated controls	

Table 4. SUMMARY OF ANIMAL STUDIES OF ERIONITE (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Erionite I (Colorado)	90% <8 μm and 6% >9.5 μm long; 85% <1 μm and 8.7% >1.4 μm diameter	male Balb/C mice	50	Intraperitoneal injection	10 mg	Single dose	Lifespan	Marked peritoneal fibrosis; peritoneal tumors in 21/42 animals	Peritoneal tumors in 6/32 animals receiving 20 mg chrysotile asbestos; no tumors in chrysotile group at 2 mg; saline controls and untreated controls had no tumors	Suzuki and Kohyama (1984)
Erionite II (Nevada)	95% <8 μm and 4% >9.5 μm length; 82% <0.5 μm and 100% <1 μm diameter	Male Balb/C mice	20	Intraperitoneal injection	0.5 mg	Single dose	Lifespan	Peritoneal mesothelioma in 6/18	Marked fibrosis in all animal groups	As above
			50	Intraperitoneal injection	2 mg	Single dose	Lifespan	Peritoneal mesothelioma in 24/44		
			75	Intraperitoneal injection	10 mg	Single dose	Lifespan	Peritoneal mesothelioma in 3/8		
Karain (Turkey) dust erionite	Not specified; also contained nonfibrous dust	Swiss albino mice	37-98/group	Intraperitoneal injection	5,10,15,20,30, or 40 mg	Single dose	Until death (up to 32 months)	Mesotheliomas in 41/321; malignant lymphoma in 31/321; mesothelioma and lymphoma in 11/321	Mesothelioma in 3/55 saline controls; lymphoma in 1/53 saline controls	Ozesmi et al. (1985)

Table 5. SUMMARY OF ANIMAL [redacted]ES ON WOLLASTONITE

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	References
Wollastonite	Not specified	Male Fischer 344 rats	Not specified	Inhalation	10 mg/m^3	12 or 24 mo	Up to 120 weeks	Results not yet available; no adverse effects on survival		AdKins and McConnell (1985)
Wollastonite (Canada)	4 samples consisting of mostly large fibers; only one sample was completely fibrous	Female Osborne-Mendel rats	20-25/ group	Intrapleural implantation	40 mg	Single dose	2 yr	Pleural tumors in 0/24, 2/25, 3/21, 5/20 animals		Stanton et al. (1981)
Wollastonite (India)	10% <2.4 μm and 90% <13 μm long; 10% <0.62 μm and 90% <2.3 μm diameter	Female Wistar rats	54	Intraperitoneal injection	20 mg	5 weekly doses	130 weeks	No peritoneal tumors; low degree of adhesion of abdominal organs	Preliminary findings only; no histological data	Pott et al. (1987b)

TABLE 6. SUMMARY OF ANIMAL STUDIES ON ATTAPULGITE

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	Reference
Attapulgite (Lebrija, Spain)	All fibers <2 μm long	SPF Fischer 344 rats	40 (20 of each sex)	Inhalation	10 mg/m^3	12 months	Lifespan	No fibrosis; peritoneal mesothelioma (1/40)	No significant excess of tumors	Wagner et al. (1987)
Attapulgite/ Palygorskite (Leicester, U.K.)	18% of fibers ≥6 μm in length and <0.2 μm in diameter	SPF F344 rats	40 (20 of each sex)	Inhalation	10 mg/m^3	12 months	Lifespan	Fibrosis; mesothelioma (3/40); malignant alveolar tumor (2/40); bronchoalveolar hyperplasia (8/40; 1 BAH with MAT); UICC cradolite produced 1/40 adenocarcinoma and 3 BAH (1 BAH with adenocarcinoma)	Some evidence of carcinogenicity	Wagner et al. (1987)
Attapulgite (U.S.)	Two samples composed entirely of short fibers of small diameters	Female Osborne-Mendel	30-50 group	Intrapleural implantation	40 mg	Single dose	2 years	Pleural tumor incidence 2/29 for both samples	Tumor incidence not statistically significant compared with controls treated with noncarcinogenic materials (17/615)	Stanton et al. (1981)
Attapulgite (French)	Mean diameter 0.06 μm; mean length 0.77 μm	Sprague-Dawley rats (sex unspecified)	Not specified	Intrapleural injection	20 mg	Single dose	2 years	No mesothelioma; UICC and Canadian chrysotile asbestos produced 19% and 48% mesothelioma incidence, respectively	Abstract only; actual data not available	Renier et al. (1987)

TABLE 6. SUMMARY OF ANI[redacted]IES ON ATTAPULGITE (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Treatment Duration	Duration of Study	Results	Remarks	Reference
Attapulgite (Lebrija, Spain)	All fibers <2 μm long	SPF F344 rats	20 of each sex	Intrapleural injection	Not specified	Single dose	Lifespan	Peritoneal mesothelioma (1/40); Pleural mesothelioma (1/40)	Tumor incidence not significant	Wagner et al. (1987)
Attapulgite (Torrejon, Spain)	0.54% of fibers (by mass) ≥6 μm in length and <0.5 μm in diameter	SPF F344 rats	20 of each sex	Intrapleural injection	Not specified	Single dose	Lifespan	Pleural mesothelioma (14/40)		As above
Attapulgite/ palygorskite (Leicester, U.K.)	18% of fibers (by mass) ≥6 μm in length and <0.2 μm in diameter	SPF F344 rats	32 (16 of each sex)	Intrapleural injection	Not specified	Single dose	Lifespan	Pleural mesothelioma 30/32; UICC Crocidolite produced 34/40 mesothelioma; chrysotile produced 19/40 pleural tumors		As above
Attapulgite (palygorskite)	37.5% <2μm long; 70% <5μm	Wistar Rats	40	Intraperitoneal injection	3 x 25 mg	Weekly	Lifespan	65% of animals developed peritoneal mesothelioma; first tumor appeared at day 275	Chrysotile asbestos produced 30-67% tumor incidence	Pott et al. (1974)
Attapulgite (France, Spain, U.S.)	Not specified; composed of the short, thin fibers	Rats	Not specified	Intraperitoneal injection	Not specified	Single dose	Not specified	No excess tumors observed with 3 types of attapulgite	No experimental details or data reported	Pott et al. (1985)
Attapulgite	mean [illegible] length <1 μm	NMRI mice	60 of each sex	Feeding	1% or 3%	25 mo	25 mo	No toxicities; no tumors		Brune and Deutsch-Wenzel (1983)

Table 7. SUMMARY OF ANIMAL ST[redacted]ON ARAMID FIBERS

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Duration of Treatment	Duration of Study	Results	Remarks	References
Ultrafine Kevlar®	90% <1.5 μm diameter; >75% less than 20 μm long	Male and female Crl:CD®BR rats	100/sex/dose group	Inhalation	0, 2.5, 25, 100, 400 fibers/mL (0.08, 0.32, 0.6 or 2.23 mg/m^3)	104 weeks for 3 low dose levels; 52 weeks for the highest dose group.	2 yr	Dose-related pathological lesions in lungs including minimal collagenized fibrosis, type II pneumocyte hyperplasia, alveolar bronchiolarization; cystic keratinizing squamous cell carcinoma in 1/36 males and 6/56 females exposed to 400 fibers/ml, and in 4/69 females at 100 fibers/ml	No positive control groups included in the study	Reinhardt (1986)
Ultrafine Kevlar®	60-70% <1 μm diameter and between 10-30 μm long	Male Crl:CD rats	20/group	Inhalation	0.1, 0.5, 3 or 18 mg/m^3	2 weeks	Up to 6 mo post exposure	Slight collagen fiber deposition by 6 mo post exposure in high dose group only (18 mg/m^3); No adverse pulmonary response at lower doses	Short-term exposure	Lee et al. (1983)
Kevlar® fibers	13% <5 μm aerodynamic diameter	Male CrR:CD rats	20	Inhalation	18 mg/m^3	2 weeks	Up to 6 mo post-exposure	No collagen formation in lung	Short-term exposure	Lee et al. (1983)
Kevlar® fibers	90% <11 μm long; 90% <0.75 μm diameter	Female Wistar rats	Not specified	Intraperitoneal injection	4x5 mg	4 weekly doses	130 weeks	Peritoneal tumors (mesothelioma/ sarcoma) in 3/53 treated with Kevlar	Preliminary results only	Pott et al. (1987b)

TABLE 7. SUMMARY OF ANIMAL STUDIES ON ARAMID FIBERS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Frequency/ Duration of Treatment	Duration of Study	Results	Remarks	References
Kevlar® pulp	Mostly aggregates of large fibers; a small proportion composed of fine fibrils; (96% of these fibrils <1 μm and 50% <0.25 μm in diameter)	Male AF/Han rats	32-48/ group	Intraperitoneal injection	0.25, 2.5 or 25 mg	Single dose	130 weeks	Minimal fibrosis; Peritoneal mesothelioma in 2/32 animals treated with 25 mg of test fibers; no mesotheliomas in low dose groups mesotheliomas (0/32 and 0/48 at 2.5 mg and 0.25 mg, respectively)	Untreated controls had no tumors (0/48)	Davis (1987)
Kevlar® polymer dust	Not specified; contained small proportion of respirable fibers (<1.5 μm diameter) and mostly large nonrespirable fibers (up to 150 μm diameter)	Rats (strain/ sex not specified)	Not specified	Intratracheal instillation	25 mg	Single dose	Up to 21 mo	Only nonspecific dust cell reactions		Reinhardt (1980)
Nomex® aramid	2-100 μm long; 2-30 μm diameter	Rats (strain sex unspecified)	Not specified	Intratracheal instillation	2.5 mg	Single dose	Up to 2 yr post treatment	Mild tissue reactions; no lung fibrosis		Reinhardt (1980)

Table 8. SUMMARY OF ANIMAL STUDIES ON CARBON FIBERS

ber pe	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Duration/ Frequency of Treatment	Duration of Study	Results	Remarks	References
N-based rbon ber	7 um diameter 20-60 μm long	Male albino rats of the CD BR sprague Dawley Strain	10-20 animals per group	Inhalation	20 mg/m^3 (16-23 mg/m^3)	4,8,12, or 16 weeks	Up to 32 weeks after 16 wk of exposure	No lung pathology	Large diameter fibers	Owen et al. (1986)
opped rbon ibers	99% nonfibrous 0.8% carbon fibers with 1-2.5 μm diameter and up up to 15 μm long	SPF guinea pigs	A total of 13 exposed animals; 2 negative controls	Inhalation	370 nonfibrous particles/ml 2.9 fibers/ml	7-104 hr	1-144 days after exposure	No pathological effects observed in animals at interval sacrifice	Dust cloud was predominantly nonfibrous; short exposure; small number of animals	Holt and Horne (1978)
opped rbon ibers	Not specified	SPF guinea pigs	2-9 animals per group	Inhalation	Not specified	7-100 hr	Up to 2 yr	No evidence of pathological changes in the lungs	Short exposure; respirable fraction was mainly non-fibrous particles	Holt (1982)
rbon st	0.2-15 μm diameter	Male and female SPF Wistar rats	12	Intraperitoneal injection	50 mg/kg (10-15 mg/ rat)	Single dose	1 or 3 mo	No evidence of fibrosis; positive control animals receiving chrysotile asbestos (2.5 mg) showed diffuse fibrosis by 3 months;	Short observation period	Styles and Wilson (1973)
rbon ibers	20% <1 μm and 35-40% <2 μm diameter	Male Fischer 344 rats	Not reported	Intraperitoneal injection/ Intratracheal instillation	Not reported	1 single i.p. dose; 2 intra-tracheal doses	Up to 2 yr	No tumors or toxicity	Preliminary oral report only	Parnell (1987)

TABLE 8. SUMMARY OF ANIMAL STUDIES O[redacted]N FIBERS (CONTINUED)

ber pe	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Duration/ Frequency of Treatment	Duration of Study	Results	Remarks	References
-lyacryl- itrile AN)-based rbon fibers	Not reported	Rats (strain, sex not specified	Not reported	Intratracheal instillation	Not reported	Single dose	1-9 mo	Both preparations of carbon fibers induced lung fibrosis; the fibrogenicity of chrysotile asbestos was several-fold higher than that of carbon fibers	No experimental details were provided	Troitskaya et al. (1984)
mixture carbon bers and specified astic	Not reported	Sprague Dawley rats (sex unspecified)	Not reported	Intratracheal injection	Not reported	Not specified	8 mo	No lung fibrosis; only moderate foreign body reaction	Experimental details and results were not not available for evaluation	Swensson (1979) (as reported by Gross, and Braun, 1984)
rbon bers	Not reported	Male & Female Sprague-Dawley rats	40/sex	Subcutaneous implantation	25 mg in a 2 cm disc	Single dose	Life span	Local sarcoma (tumor incidence not specified)	Details of findings not available	Maltoni et al. (1982b, 1987)
rbon ber lament	1.5 cm long or 5 cm long; diameter not specified	Wistar rats (sex not specified)	10 or 50 rats/ group	Intramuscular implantation	Not specified	Single dose	18 mo	No malignant change	Test materials were either nonfibrous or large filament; route of exposure not relevant to evaluation of of lung carcinogenicity	Tayton et al. (1982)
rbon n fibrous wder	particle size not specified	Wistar Rats	50 rats	Intramuscular implantation	Not specified	Single dose	18 months	No malignant change		

TABLE 8. SUMMARY OF ANIMAL STUDIES [redacted]ON FIBERS (CONTINUED)

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose/ Concentration	Duration/ Frequency of Treatment	Duration of Study	Results	Remarks	References
Carbon fiber reinforced carbon	7 μm diameter; 20–100 μm long	CHBB:CH rabbits (sex not specified)	8 per group	Intramedullary implantation	50 mg	Single dose	2 or 12 weeks	Small amount of fibrosis around some carbon fibers	Method of exposure not relevant to the determination of the potential induction of lung toxicity and fibrosis	Neugebauer et al. (1981)
Ground carbon fibers of 4 types	Not specified	Male C3H/HeJ mice	40 per group	Skin painting	2.5 mg (25 μL of 10% w/v fiber suspension)	3 times weekly	Lifespan		In the CF treated group, neither tumor incidence nor time to onset of tumors was significant compared to benzene-treated control animals (0/40) in this study but were judged to be weakly oncogenic using historical benzene controls (0/285); positive controls receiving 0.1% methylcholanthrene had increased incidence of squamous cell carcinoma and papilloma (33/38)	DePass (1982)
(a) continuous filament (CF)-pitch-based								Skin tumors at site of application; squamous cell carcinoma (1/40); papilloma (1/40)		
(b) short-fibered pitch-based (MAT)								Fibrosarcoma (1/40); hemangiosarcoma (1/40)		
(c) polyacrylonitrile (PAN)-based								No tumors (0/40)		
(d) oxidized PAN-based								Leiyomyosarcoma (0/40)		

TABLE 9. SUM[redacted] ANIMAL STUDIES ON POLYOLEFIN FIBERS

Fiber Type	Fiber Dimension	Species	Number of Animals	Route of Administration	Dose of Concentration	Frequency/ Duration of Treatment	Duration of Study	Results	Remarks	References
Polypropylene	90% <2.1 μm and 50% <1.1 μm diameter; 90% <23 μm and 50% <7.4 μm length	Female Wistar rats	53	Intraperitoneal injection	5 x 10 mg	Weekly injection for the first 5 weeks	Lifespan	Peritoneal tumors (mesothelioma/ sarcoma) in 2/53 animals	Preliminary results only	Pott et. al (1987b)
Polyethylene dust	3.75 μm diameter	Male and female albino Wistar rats	6 of each sex	Intraperitoneal injection	50 mg/kg (10-15 mg/kg animal)	Single dose	Sacrificed at 1 or 3 mo after treatment	No tumors or fibrosis; diffuse fibrosis observed in chrysotile treated animals	Short observation period; small number of animals	Styles and Wilson (1973)
Polypropylene dust	4-50 μm diameter	As above	As above	As above	As above	As above	As above	As above		
Ozonized Polyethylene SHFF Ozonized Polypropylene SHFF HFF Polypropylene	Not specified	Male Long-Evans Rats	40/group	Intratracheal insufflation	Not specified	Single dose	21 mo	No lung tumors or fibrosis in any treated groups	Limited study; No data available on the characteristic on the test materials, dose and details on the method of administration	MB Research Laboratories (1980)