

Review Article

# Man-Made Respirable-Sized Organic Fibers: What Do We Know about Their Toxicological Profiles?

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**Abstract:** Man-made organic fibers (MMOFs) have been manufactured for over 50 years. Until recently, there have been few concerns raised regarding the safety of organic fiber dusts. This is due, in large part, to the perception that the dimensions of most, if not all, of these products were too large to be inhaled into the distal lungs of workers, i.e., were considered to be nonrespirable. A brief review of some of the issues related to organic fiber toxicology is presented herein. Some of the organic fiber-types used in commerce are identified and some fundamental tenets of fiber toxicology are discussed. In addition, the European Union, in their recent consideration for banning chrysotile asbestos fibers, evaluated some organic fiber substitutes and compared them to the hazards of asbestos. A brief review of their conclusions is described below. Finally, the results of some recent studies assessing the mechanisms of biodegradability of para-aramid respirable-sized, fiber-shaped particulates (RFP) are presented. Para-aramid (p-aramid) RFP are the most extensively-studied respirable organic fiber-type and RFP is the new term which describes respirable-sized organic fibers (ECETOC, 1996) (1). The results of these studies provide clues regarding the mechanism(s) of p-aramid RFP shortening in the lungs of exposed animals, and may be relevant for humans.

**Key words:** Man-made organic fibers (MMOF), Respirable-sized fiber-shaped particulates (RFP), Para-aramid, P-aramid, Cellulose, Polyvinyl alcohol fibers

## Introduction

Synthetic man-made organic fibers (MMOFs) have been in production for a period greater than 50 years. Despite the length of time in commerce, there is a paucity of information regarding the pulmonary toxicological effects of inhaled MMOF fibers and dust<sup>1</sup>. This is due, in large part, to the perception that: 1) exposures to airborne MMOFs generally were considered to be very low in occupational settings; and 2) exposures to MMOF fibers or dust in the workplace was assumed to be non-respirable (i.e., it was assumed that airborne MMOFs would be too large to penetrate to the distal (gas exchange regions) lung. MMOFs generally are produced as

continuous filaments in the manufacturing process and the filaments are processed in diameter dimensions that exceed the respirable range (i.e.,  $>3 \mu\text{m}$  diameter). Thus, it is assumed that if inhaled, the larger MMOF filaments would be trapped in the upper airways, wherein clearance is generally considered to be relatively rapid. However, during certain manufacturing processes, especially those involving chopping or grinding, aerosols of smaller-sized, respirable MMOFs can be formed. Moreover, in recent years, new application and production methods (i.e., microfibers and flocking material) have been developed by the MMOF industry and these modifications have resulted in reduced fiber dimensions. As a consequence, a concern has developed, that in certain occupational environments, workers may be exposed to airborne respirable MMOFs<sup>1,2</sup>.

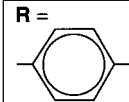
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As with synthetic vitreous fibers and asbestos fibers, it is generally regarded that the toxicity of inhaled fibrous materials is governed, in large part, by the 3 factors, often known as the three “D”s—namely, dose, dimension and durability. These concepts can be described by the following: 1) Dose—this refers to the numbers of inhaled fibers which deposit in the alveolar regions of the lung over time and generally is correlated with the development of pulmonary toxic effects. (b) Dimension—refers to the idea that thinner fibers and longer fibers are more toxic to pulmonary cells and generally more persistent in the lung (fibers with lengths  $>15 \mu\text{m}$  are not as efficiently cleared from the lung by alveolar macrophages; (c) Durability—the severity of pulmonary adverse effects in animals inhaling inorganic fibers is proportional to the biopersistence (or durability) of the retained fiber in the lung; biopersistence is determined by both fiber length (longer = more biopersistent) and chemical durability (resistance to degradation in lung fluids or pulmonary cells)<sup>2</sup>. One could consider a 4th D as a factor in fiber toxicity. This “D” would stand for—distribution and

would describe the translocation pattern of an inhaled fiber following deposition. For instance, a fiber (such as chrysotile asbestos) is likely to be more hazardous if, following deposition into the alveolar regions of the lung, it translocates through alveolar epithelial cells into the pulmonary interstitium. In contrast, other fiber-types may remain in the alveolar parenchymal environment and thus interact primarily with alveolar macrophages.

Polymeric man-made organic fibers are synthesized from organic polymers that are derived from petroleum-based chemicals. Some examples of MMOF are: polyamides (nylon, aramid), polyester, polyolefins (polyethylene, polypropylene), and polyvinyls (Table 1). Petroleum-based MMOFs have been utilized in the production of textiles for carpets, clothing, bedding, curtains, draperies, and upholstery. In addition, some MMOF fiber-types are used for tire cords (e.g., p-aramid, nylon), protective clothing (e.g., m-aramid) industrial fabrics, ropes and cables, and friction materials (brake pads and gaskets).

**Table 1. Examples of Semi-Synthetic and Man-Made Organic Fibers**

| <b>Semi-Synthetic Fibers</b>  |   |
|---|---|
| (Derived from Natural Plant Fiber, Cellulose)                                       |   |
| Regenerated cellulose derivatives   |   |
| Viscose rayon   |   |
| Cuprocellulose  |   |
| Cellulose acetate   |   |
| Cellulose triacetate  |   |
| Solubilized cellulose derivative  |   |
| Lyocel  |   |
| <b>Man-Made Polymeric Organic Fibers</b>  |   |
| Polymer   | Monomer   |
| Polyamides  |   |
| Aliphatic (nylon)   | $-\text{NH}-\text{CO}-(\text{CH}_2)_4-\text{CO}-\text{NH}-(\text{CH}_2)_5-$ |
| Aromatic (aramid, meta- & para)   | $-\text{NH}-\mathbf{R}-\text{NH}-\text{CO}-\mathbf{R}-\text{CO}-$           |
| Polyester   |   |
| Polyethyleneterephthalate   | $-\text{O}-\text{CO}-\mathbf{R}-\text{CO}-\text{O}(\text{CH}_2)_n-$         |
| Polyolefins   |   |
| Polyethylene  | $-\text{CH}_2-\text{CH}_2-$   |
| Polypropylene   | $-\text{CH}(\text{CH}_3)-\text{CH}_2-$                                      |
| Polyvinyls  |   |
| Polyacrylonitrile (orlon)   | $-\text{CH}(\text{CN})-\text{CH}_2-$  |
| Polyvinylchloride   | $-\text{CH}(\text{Cl})-\text{CH}_2-$  |
| Polyurethane (Elastane)   | $\text{HOR}-\text{OCO}-\text{NH}-\mathbf{R}-\text{NH}-\text{COO}-$          |
|  |   |

## European Union Consideration of a Ban on Chrysotile Asbestos and Evaluation of Substitute Organic Fiber-types

The European Union recently considered a ban on the use of chrysotile asbestos fibers. Such a consideration requires an evaluation of the health effects of substitute materials. The main organic fiber substitutes that were evaluated for the residual uses of chrysotile were para-aramid (p-aramid), polyvinyl alcohol (PVA), and cellulose fibers. Much of the risk consideration was based upon hazard assessment, including fiber dimensions and durability (as discussed above) and potential exposure assessment, i.e., the likelihood that respirable fiber aerosols would be generated<sup>3</sup>. Here is a brief summary of their conclusions:

### *Polyvinyl alcohol (PVA) fibers*

It was concluded that the diameter of most manufactured PVA fibers is approximately 10–16  $\mu\text{m}$ , and therefore exceeds the criteria for respirable size (i.e., generally considered to be 3  $\mu\text{m}$  for fiber counting purposes). There is evidence that PVA fibers can fibrillate, however, most of the dust particles measured in the working atmosphere appear to be of a nonfibrous particulate nature. Although the toxicological data base for PVA fibers is sparse, the results of generally negative epidemiology and exposure assessment studies indicate that, relative to asbestos, there are significantly reduced human exposures to PVA fibers<sup>3</sup>. Morinaga and colleagues recently conducted a retrospective cohort study of male workers exposed to PVA fibers. A total of 447 exposed and 2416 non-exposed male workers were evaluated. Lung cancer SMR rates were 0.86 for the workers with 20 or more years of employment. The authors concluded that there was no difference in lung cancer risk between workers exposed to PVA fibers when compared to non-exposed workers<sup>4</sup>.

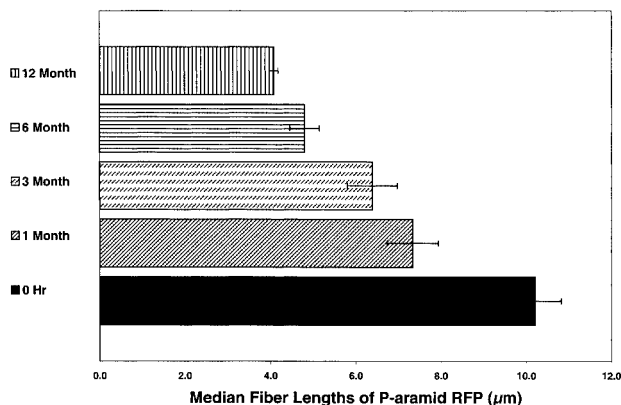
### *Para-aramid fibers and RFP*

Para-aramid (p-aramid) RFP form the respirable component of para-aramid fibers in pulp, and generally are used in friction products such as gaskets and brake linings. As discussed above, RFP is an acronym for respirable-sized, fiber-shaped particulates and is the new nomenclature used for identifying respirable-sized organic fibers<sup>1</sup>. Para-aramid RFP (also referred to herein as fibrils) generally have diameter dimensions of 0.3–0.7  $\mu\text{m}$  and are produced under conditions of abrasion of p-aramid fibers, which are considered to be nonrespirable, having diameters in the range of 12–15  $\mu\text{m}$ <sup>5</sup>. Recent inhalation toxicity studies with rats comparing the biopersistence of p-aramid RFP with asbestos fibers have

demonstrated that the fibrils were less biopersistent than chrysotile asbestos. The longer p-aramid RFP were shortened in the lungs of rats while the longer chrysotile asbestos fibers were preferentially retained. Significant differences were also measured in cell proliferation parameters, with p-aramid producing a transient increase in terminal bronchiolar cell proliferative responses, while chrysotile asbestos fibers produced a sustained cell proliferative response in the airways, lung parenchyma, and subpleural regions of the lung<sup>6</sup>. Exposure studies have recently been reported by Cherrie *et al.*<sup>7</sup> who sampled para-aramid process workers in the United Kingdom. Exposure levels, as expressed as the geometric mean of the time-weighted average ranged from 0.005 to 0.4 f/ml, with 95% of all measurements <0.2 f/ml.

### *Cellulose fibers*

Cellulose fibers are produced from natural sources and have historically been used in the paper industry. Although epidemiological data for cellulose is sparse, there appears to be little evidence of disease in workers, even at high exposure levels in the workplace<sup>3</sup>. Exposures to hardwood-associated wood dust is linked with development of sinonasal cancer; however, exposure to softwoods seem to be significantly less potent in producing similar effects, indicating that cellulose was not the causative agent. With regard to experimental studies, Muhle and colleagues recently reported that cellulose RFP were more biopersistent than chrysotile in the lungs or rats, however, this pulmonary toxicokinetic study likely was conducted at overload concentrations<sup>8</sup>. There exists a paucity of data regarding the pulmonary toxicity of inhaled cellulose RFP. Harrison *et al.* noted that there exist surprisingly few data on cellulose despite the fact that this fiber is associated with products with rather wide commercial applications<sup>3</sup>. The few *in vitro* and *in vivo* experimental studies that have been conducted suggest that cellulose RFP may be biopersistent in the lung and may produce pulmonary inflammation<sup>8–10</sup>. Cullen and coworkers recently conducted a 3-week inhalation study with one form of cellulose RFP (i.e., mechanical wood pulp) at 1000 f/ml<sup>11</sup>. It was reported that cellulose induced an early pulmonary inflammatory response in rat lungs, as assessed by bronchoalveolar lavage, which peaked at 1 day following the start of inhalation and thereafter declined. These investigators concluded that the cellulose material studied was less inflammogenic than crocidolite and that the extent of the inflammatory response within the lung was reduced with continued inhalation exposure<sup>11</sup>. It is unclear whether the type of cellulose utilized in these studies is a representative form of cellulose that is widely used in the paper industry. In summary, the few *in vitro* and *in vivo* studies that have been



**Fig. 1.** A bargraph from an earlier study (Warheit *et al.*, 1994) demonstrating progressively reduced median lengths of retained p-aramid RFP with increasing transit time in the lungs of rats following 2-week high dose inhalation exposures.

The progressive reduction in lengths represents cleavage of the p-aramid fibrils in the lungs of exposed rats.

conducted suggest that cellulose RFP may be biopersistent in the lung and may produce pulmonary inflammation<sup>8-11</sup>).

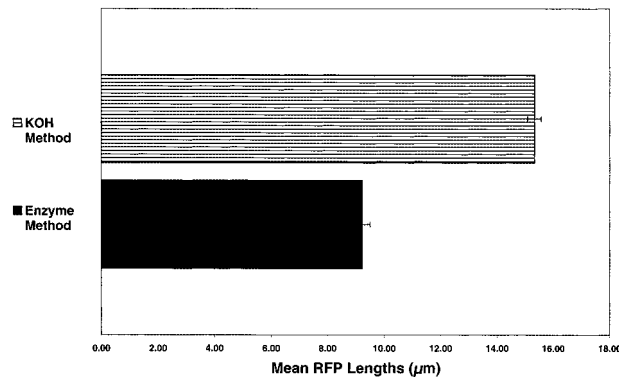
Since cellulose represents a family of materials, there is a great need to assess the toxicity of the various respirable forms of this organic fiber-type.

The European Commission Health and Consumer Protection Directorate Scientific Committee concluded that chrysotile asbestos fibers are fundamentally more hazardous than p-aramid, PVA, or cellulose fibers. As a consequence, they have endorsed the opinion that the continued use of chrysotile asbestos in cement products and friction materials is not justifiable, given the availability of less hazardous organic fiber substitutes<sup>3</sup>).

### Experimental Studies with Para-Aramid RFP

Para-aramid RFP are one of the very few synthetic respirable organic fibre-types that have been extensively evaluated in inhalation toxicity studies, and the pulmonary toxicity and biopersistence of this material in the lungs of exposed rats and hamsters has been investigated.

The results from several acute and chronic inhalation toxicology studies in rats have demonstrated that p-aramid RFP have low biopersistence, i.e., break down transversely into smaller fragments in the lungs of exposed rats<sup>12-16</sup>) (see Fig. 1). Similarly, results from inhalation studies with p-aramid exposed hamsters have confirmed these data in another species<sup>17</sup>). The finding of retained p-aramid RFP cleavage over time in the lungs of two rodent species<sup>12, 13, 17</sup>) along with the lack of lung or serosal tumors in rats following chronic



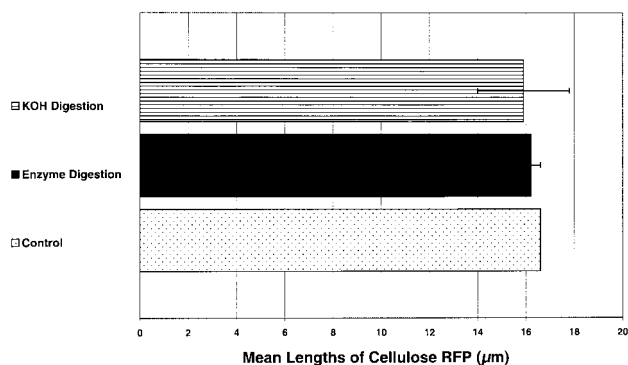
**Fig. 2.** Bar graph summarizing mean lengths of p-aramid fibrils after recovery from the lungs of exposed rats.

Digestion of lung tissue after enzyme or KOH digestion treatment. The lungs of rats instilled with p-aramid RFP were digested using one of the two digestion methods 24 hours postexposure. The mean lengths of p-aramid RFP from the enzyme treated solution were significantly shorter than those RFP recovered from the lungs of KOH-treated animals (\* $p < 0.05$ ), suggesting that the interactions of p-aramid RFP with lung fluids followed by enzymatic digestion may account for biodegradability of p-aramid RFP.

inhalation or intraperitoneal injection studies have been utilized by IARC in 1997 as justification for supporting a Category 3 classification (Inadequate evidence for carcinogenicity)<sup>5</sup>).

The present studies were undertaken to elucidate mechanisms of p-aramid biodegradability in the lungs of exposed animals. Based on preliminary findings by Searl and Cullen<sup>18</sup>), who reported that an enzymatic digestion method artificially shortened p-aramid RFP *after* instillation into the lungs, we have postulated that 1) inhaled p-aramid RFP are biodegraded in the lungs of exposed animals via an enzymatic mechanism; and 2) this process of biodegradation is catalyzed or activated by interactions of the fibril with lung fluids prior to enzymatic cleavage.

To test this hypothesis, p-aramid RFP or cellulose RFP were instilled into the lungs of rats and the lungs were digested 1 day after exposure using two different lung digestion techniques, 1) a conventional ethanolic KOH procedure, or 2) an enzymatic method (designed to simulate lung enzymes). Our results demonstrated that the enzyme method but not the KOH method artificially reduced the lengths of p-aramid RFP recovered from rat lungs (Fig. 2). In contrast, neither the enzyme or KOH procedure affected the length dimensions of cellulose RFP (Fig. 3)<sup>19</sup>). Complementary *in vitro* noncellular studies were carried out wherein p-aramid or cellulose RFP were incubated with lavaged fluids from a normal rat and then underwent simulated digestion procedures with either KOH or enzyme digestion preparations. The lengths of p-



**Fig. 3.** Mean lengths of cellulose RFP following recovery from the lungs of exposed rats.

Digestion of lung tissue following enzyme or KOH digestion treatment. The lungs of cellulose RFP-instilled rats were digested using one of the two methods 24 hours postexposure. The mean lengths of cellulose RFP from the enzyme treated solution were not significantly reduced when compared with RFP recovered from the lungs of KOH-treated animals, suggesting that the data obtained with p-aramid in Fig. 2 are not nonspecific.

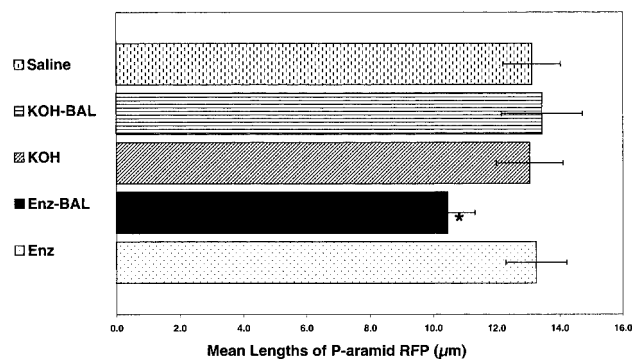
aramid RFP were decreased when incubated with BAL fluids and then processed via the enzyme digestion method but not with KOH, thus confirming the *in vivo* results (Fig. 4). In contrast, no change in cellulose RFP lengths were measured following incubation with BAL fluids and subsequent processing with either KOH or enzyme method (Fig. 5)<sup>19</sup>.

In another set of *in vitro* cellular experiments, cultures containing rat lung epithelial cells, alveolar macrophages, or co-cultures of epithelial cells and macrophages were incubated with p-aramid fibrils for periods of 1 hour, 1 day, or 1 week to assess whether RFP cleavage occurs directly in the phagocytic cells. The lengths of fibrils were measured using scanning electron microscopy methods. The results demonstrated that 1) no cleavage was measured in fibrils exposed to the epithelial cell cultures at any time point; however, cleavage of p-aramid RFP was measured at 1 day and 1 week postexposure in the macrophage and co-cultures (Fig. 6)<sup>19</sup>.

The conclusions derived from these studies strongly suggest that components of lung fluids coat and activate the p-aramid RFP as a prerequisite for enzymatic cleavage. This process could play a significant role in facilitating the transverse cleavage or shortening of inhaled p-aramid RFP in the lungs of exposed rats and hamsters.

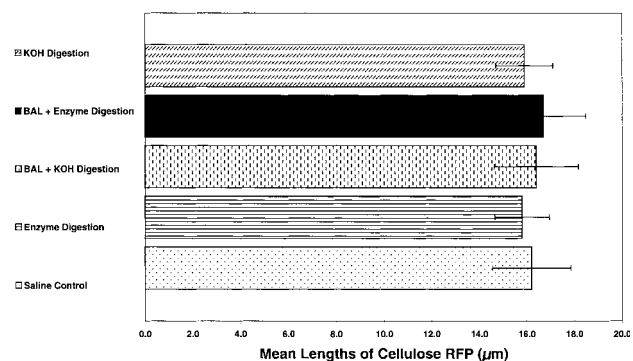
## Acknowledgments

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**Fig. 4.** BAL fluids activate para-aramid RFP for biodegradation and (subsequent enzymatic cleavage).

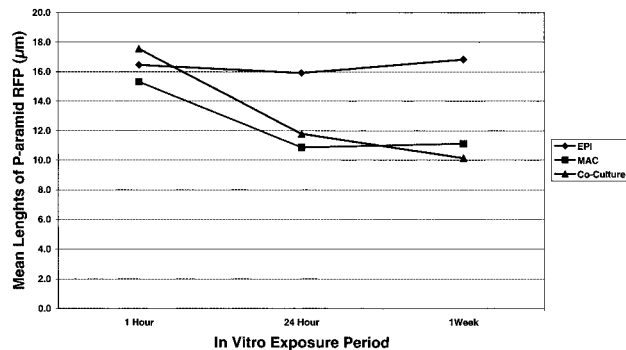
A preparation of p-aramid RFP was incubated in bronchoalveolar lavage fluids or saline for 3 hrs and then either processed with the KOH digestion technique or an enzyme technique. The findings indicate that the combination of incubation in BAL fluids followed by digestion by the enzymatic technique resulted in a decrease in measured RFP lengths (Enz-BAL) when compared to the other treatments. The results of this experiment could simulate a possible mechanism that occurs in the lungs of exposed animals. \*  $p < 0.05$ .



**Fig. 5.** A preparation of cellulose RFP was incubated in bronchoalveolar lavage fluids or saline for 3 hrs and then either digested with the KOH digestion or an enzyme method.

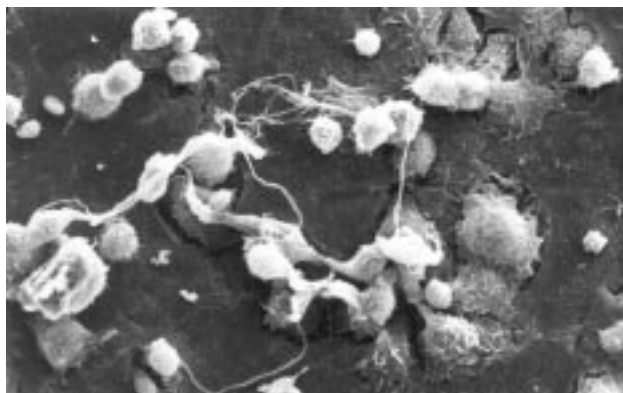
The results indicate that the combination of incubation in BAL fluids followed by simulated digestion by the enzyme technique resulted in no reduction in measured RFP lengths compared to the other controls. These results demonstrate that the shortening of the p-aramid RFP demonstrated in Fig. 4 are not nonspecific and did not occur with cellulose RFP.

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**Fig. 6.** Cellular *in vitro* studies demonstrating biodegradability of p-aramid RFP.

Data from these *in vitro* cellular studies with p-aramid RFP demonstrated that after one hour there was no evidence of changes in the mean fibril lengths among the epithelial (EPI), alveolar macrophage (MAC) or epithelial-macrophage co-culture groups (Co-culture). However, after one day of exposure, the mean lengths of RFP exposed to the macrophage and co-culture groups were statistically different from the mean RFP lengths of the epithelial group. The results after one week were identical to the results after one day. \* $p < 0.05$  when compared to corresponding EPI culture at each time period.



**Fig. 7.** A scanning electron micrograph is presented which is representative of epithelial-macrophage cell cultures exposed to p-aramid RFP after 1 day. magnification = 400 $\times$ .

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