WHAT'S NEW IN FIBER ANALYSIS OF HUMAN TISSUE SAMPLES?

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Asbestosis

Our database includes 170 patients with asbestosis for whom fiber analyses of lung tissue samples have been performed (Table 1). Lung cancer was also present in 74 of these cases, pleural mesothelioma in 36, and peritoneal mesothelioma in 9. The median asbestos body count by light microscopy (LM) is 22,000 per gram of wet lung, and the median uncoated fiber count by scanning electron microscopy (SEM) for fibers 5 μm or greater in length was 179,000 fibers/gm. The median count is higher for patients with peritoneal mesothelioma. The main fiber type identified was amosite.

There is a linear and statistically significant correlation between the concentration of fibers 5 μm or greater in length as determined by SEM and the severity of pulmonary asbestosis as determined histologically, although there is a wide range of scatter of the data points. Cases lacking histological criteria for asbestosis (peribronchiolar and/or alveolar septal fibrosis *and* asbestos bodies in histologic sections) have a fiber content below the 95% confidence interval for bona fide cases of asbestosis. Therefore, patients with clinically significant interstitial fibrosis but without asbestos bodies in histologic sections are unlikely to have asbestosis. There appears to be a threshold for the development of asbestos-related pulmonary parenchymal fibrosis.

Mesothelioma

Our database includes 281 patients with malignant mesothelioma for whom fiber analyses of lung tissue samples have been performed (Table 2). Pleural mesothelioma was present in 263 cases and peritoneal mesothelioma in 18. The median asbestos body and uncoated fiber count is greater for those patients with asbestosis as compared to those with plaques alone, and the content for patients with plaques is greater than those with neither plaques nor asbestosis. Patients with peritoneal mesothelioma tend to have higher counts than those with pleural mesothelioma. Elevated tissue asbestos content was found in 89% of cases. The remaining 11% appear to be spontaneous, idiopathic or background cases. The main fiber type identified for the group as a whole was amosite.

Roggli et al. studied 1445 cases of mesothelioma with respect to exposure history and found that 94% of cases fell into one or more of 19 exposure categories, including 12 industries, six occupations, and one non-occupational exposure group (Table 3). Four industries accounted for more than 100 cases each: shipbuilding, U.S. Navy/merchant marine, construction, and insulation industries. Two occupations accounted for more than 100 cases each: pipefitter/welder and boilermaker. Exposure in more than one category was identified in 21% of cases. The distribution of occupations was similar to that reported by the Australian Mesothelioma Registry.

The one non-occupational group with a significant number of cases was household contacts of asbestos workers, accounting for 6% of mesothelioma cases. More than half of mesotheliomas in women occurred in individuals with a reported exposure as a household contact. Fiber analyses showed an elevated fiber content in about 70% of these cases, and the concentrations were generally equivalent to a mild to moderate occupational exposure. An elevated asbestos content was found in about 70% of all women with pleural mesothelioma, but less than half of those with peritoneal mesothelioma.

Epidemiologic studies have shown a strong relationship between exposure to commercial amphibole fibers and mesothelioma risk. In experimental animal studies, long fibers (5 μm or greater in length) are associated with carcinogenic potential. In addition, levels of amphibole fibers 5 μm or greater in length correlate best with disease risk in cohorts of mesothelioma patients. These findings in turn relate to the biopersistence of amphibole fibers in lung tissue relative to chrysotile.

Further elucidation of the role of long commercial amphibole fibers in the pathogenesis of mesothelioma has come from the work of Boutin et al. These investigators have found that black spots on the parietal pleura tend to concentrate long thin amphibole fibers. These black spots correlate with the location of stoma of lymphatics that drain the pleural space, and the size of these stomata is such that long thin commercial amphibole fibers are likely to be retained there. This is also the location where mesothelioma is thought to develop. Other investigators have also found long commercial amphibole fibers in pleural tissues. Although the studies of Suzuki et al have tried to implicate short chrysotile fibers in the visceral pleura and within mesothelial tumors themselves, these studies are confounded by possible fiber contamination. Furthermore, they are not consistent with the epidemiologic findings nor with the animal studies regarding short fibers. More on the Suzuki paper will be said at the DRI meeting in Miami in November.

Benign Asbestos-Related Pleural Disease

Our database includes 197 patients with benign asbestos-related pleural disease for whom fiber analyses of lung tissue samples have been performed (Table 4). Mesothelioma was also present in 88 of these cases and lung cancer in 54. The median asbestos body count for patients with parietal pleural plaques was 680 asbestos bodies per gram, and the median uncoated fiber count by SEM was 19,600. These values are far lower than we have observed in patients with asbestosis. The values for patients with rounded atelectasis were almost identical. Elevated fiber content was found in 95% of our cases of pleural plaques. The predominant fiber type was amosite.

Fiber Burdens by Occupational Exposure Category

The results of fiber analysis are displayed according to exposure category in Table 5. Insulators have by far the highest fiber burdens, and two thirds have asbestosis. This group no longer includes pipefitters, boilermakers, and plasterers (See 1st Edition). Individuals in asbestos manufacturing have the next highest values, followed by shipyard workers, power plant workers, molten metal workers, U.S. Navy/merchant marine, construction workers, oil and chemical refinery workers, railroad workers, and brake repair workers. Household contacts of asbestos workers have levels in between those of power plant workers and molten metal workers. The values in this table differ from those of the study of 1445 mesothelioma cases because diseases in addition to mesothelioma are included. Building occupants in general have levels that are not distinguishable from those of a reference population, with a few exceptions.

We have separately reported our findings in 10 auto mechanics with pleural mesothelioma. These patients either had lung fiber burdens indistinguishable from a reference population (about half of the cases) or had elevated levels of commercial amphibole fibers (mostly amosite). Epidemiologic studies have failed to demonstrate an increased risk of mesothelioma among auto mechanics. The nature of brake dust and experimental studies showing the lack of pathogenicity of short fibers (< 5 μm in length) lend further support to the innocuous nature of friction product exposures.

Fiber Type

The results of the analyses of more than 11,000 fibers from more than 550 cases are shown in Table 6. The main fiber type accumulating in lung tissue samples across all disease and exposure categories is amosite asbestos. Tremolite is the next most common fiber type. Some investigators have claimed that tremolite is removed from chrysotile during processing. The results of our analyses of end-users and those of Churg involving end-users as well as chrysotile miners and millers do not support this claim. The likely

source of the tremolite that we have identified is contamination of Canadian chrysotile. Talc is also a contributing factor. Vermiculite is a possible but less likely source. Non-asbestos mineral fibers are commonly found during analysis of lung tissue samples, but these have not been implicated in asbestos-associated diseases.

Many investigators (J.C McDonald, Fred Pooley, John Craighead, J.C. Wagner, and myself) believe that the fibers that accumulate within lung tissue are those that are responsible for disease. Other investigators have suggested that chrysotile, which is more readily cleared from the lungs, may cause injury prior to its removal (hit and run theory). There is actually very little if any scientific support for this theory. Experimental studies have shown that when chrysotile asbestos related injury was present, fibers were present in excess amounts within the tissues. Furthermore, studies with man-made mineral (vitreous) fibers have shown that those fibers that persist are the ones that cause fibrosis and neoplasia.

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TABLE 1. Asbestos Content of Lung Tissue in 170 Cases of Asbestosis[a]

	N	AB/gm (LM)	N	UF/gm (SEM)
Asbestosis[b]	52	17,100	48	152,000
Asbestosis plus lung cancer	74	28,600	71	180,000
Asbestosis plus peritoneal mesothelioma	9	140,000	9	380,000
Asbestsis plus pleural mesothelioma	35	14,900	36	77,800

a. Asbestos bodies per gram of wet lung tissue as determined by light microscopy (LM) and uncoated fibers 5 μm or greater in length per gram of wet lung tissue as determined by scanning electron microscopy (SEM). Values reported as median.
b. Cases of asbestosis with neither lung cancer nor mesothelioma.

TABLE 2. Asbestos Content of Lung Tissue in 281 Cases of Mesothelioma[a]

	N	AB/gm (LM)	N	UF/gm (SEM)
Pleural Mesothelioma				
Asbestosis[b]	35	14,900	36	77,800
PPP	85	900	79	24,900
Other	143	97	137	18,900
Peritoneal Mesothelioma				
Asbestosis	9	140,000	9	380,000
PPP	3	1450	2	122,000
Other	6	10	6	6520

a. Asbestos bodies per gram of wet lung tissue as determined by light microscopy (LM) and uncoated fibers 5 μm or greater in length per gram of wet lung tissue as determined by scanning electron microscopy (SEM). Values reported as median.

PPP = parietal pleural plaques; Other = cases with neither asbestosis nor plaques (or uninformative cases with regards to plaques or asbestosis)

TABLE 3. Exposure Categories in 94% of 1445 Cases with Malignant Mesothelioma[a]

Industry	*Occupation*	*Non-occupational Exposure*
Shipbuilding	Pipefitter/welder	Household contact
U.S. Navy/merchant marine	Boiler worker	
Construction	Maintenance	
Insulation	Machinist	
Oil and chemical	Electrician	
Power plant	Sheet metal worker	
Railroad		
Automotive		
Steel/metal		
Asbestos manufacture		
Paper mill		
Ceramics/glass		

Modified from Reference 10.

TABLE 4. Asbestos Content of Lung Tissue in 197 Cases of Benign Asbestos-Related Pleural Disease[a]

	N	AB/gm (LM)	N	UF/gm (SEM)
PPP + Mesothelioma	88	970	81	25,100
PPP + Lung Cancer	54	710	54	19,200
PPP (Other)	43	330	40	14,800
Rounded Atelectasis	8	680	9	22,100

Asbestos bodies per gram of wet lung tissue as determined by light microscopy (LM) and uncoated fibers 5 µm or greater in length per gram of wet lung tissue as determined by scanning electron microscopy (SEM). Values reported as median.

PPP = parietal pleural plaques; Other = cases with neither mesothelioma nor lung cancer. Cases with asbestosis excluded.

TABLE 5. Asbestos Content of Lung Tissue by Exposure Category*

	N	AB/gm (LM)	UF/gm (SEM)
Asbestos insulator	68	55,600	342,000
Asbestos manufacturing	13	3090	104,000
Shipyard worker (other than insulator)	114	2560	38,600
Power plant worker	16	860	34,000
Household contact	28	260	24,300
Molten metal worker	13	260	21,800
U.S. Navy/merchant marine	34	220	17,400
Construction worker	44	220	14,000
Oil/chemical refinery	22	180	17,100
Railroad worker	20	55	15,900
Brake repair worker	23	19	9710
Bldg. occupant	10	1.7	13,000
Reference population	19	2.9	3100

Data are presented as median values of asbestos bodies per gram of wet lung as determined by light microscopy or uncoated fibers 5 µm or greater in length per gram of wet lung as determined by SEM. N represents the number of cases in each category.

TABLE 6. Energy Dispersive X-ray Analysis of 11,052 Fibers from 576 Cases[a]

	N	AC	TAA	Chrys	NAMF
Asbestosis[b]	48	157,000	11,000	8820	18,700
Pleural Mesothelioma					
Asbestosis	35	94,500	6700	6060	13,400
PPP	79	14,500	2540	1100	7000
Other[c]	135	3300	2000	700	7220
Peritoneal Mesothelioma					
Asbestosis	9	533,000	25,200	25,200	31,500
PPP	2	121,000	5300	16,000	21,400
Other[c]	6	300	1190	300	5340
Lung Cancer					
Asbestosis	70	252,000	15,700	11,400	22,800
PPP	52	9410	1590	1030	5870
Other[c]	121	2640	1580	850	6000
Reference Population	19	300	300	300	2830

a Values reported are the median of fibers 5 μm or greater in length per gram of wet lung tissue as determined by SEM.
b Asbestosis cases without mesothelioma or lung cancer.
c Other represents cases with neither asbestosis nor plaques (or uninformative cases).
AC = amosite and crocidolite; Chrys = chrysotile; NAMF = non-asbestos mineral fibers; PPP = parietal pleural plaques without asbestosis; TAA = tremolite, anthophyllite, and actinolite.