Mineral Fiber Content of Lungs in Patients with Mesothelioma Seeking Compensation in Québec

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Asbestos fibers (AF) and ferruginous bodies (FB) in lung parenchyma from 50 workers seeking compensation from the Workers' Compensation Board of Québec for pleural or peritoneal mesothelioma were analyzed using transmission electron microscopy (TEM) equipped with energy-dispersive spectrometer (EDS) and phase-contrast microscopy (PCM). These workers had been occupationally exposed in mining and milling activities (12 were from Asbestos Township and 11 from Thetford Mines) and 27 were from other types of industry (asbestos factory, shipyard, etc.). For comparison, analyses of lung tissue at autopsy were done in a group of 49 subjects from a reference population. A 95% confidence interval upper limit of 540 AF < 5 μm/mg and a 95% confidence interval upper limit of 161 AF ≥ 5 μm/mg dried lung tissue were found for the reference population. Similarly, a concentration of FB of 142 FB/g constituted the upper limit of detectable FB in the lungs of the reference population. Forty-eight of the 50 workers with mesothelioma had either a ferruginous body or total asbestos fiber count greater than the 95% confidence interval for the reference population; the remaining two had amosite and/or crocidolite concentrations greater than the 95% confidence interval for the reference population. The fiber types were different in the three groups, with the lungs of workers from Thetford Mines containing only chrysotile and tremolite, those from Asbestos Township containing chrysotile, tremolite, amosite, and crocidolite, and those in other industries containing largely amosite and crocidolite. We conclude that in this population of workers seeking compensation for mesothelioma, fiber analysis confirmed occupational asbestos exposure in every case. The fiber types responsible for the tumors are probably different in the three different groups. Dufresne A, Bégin R, Churg A, Massé S. Mineral fiber content of lungs in patients with mesothelioma seeking compensation in Québec.

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In the assessment of workers with mesothelioma for possible workers' compensation, it is imperative to have information on past exposure to establish a link between work and disease.

Whereas a detailed work history may provide appropriate information in most cases, estimates of past occupational exposure to airborne contaminants such as dust particles may be difficult and at times impossible to obtain. Accurate records of specific job titles may not be available; direct industrial hygiene measurements of current exposures may not be representative of past occupational exposure to the substances of interest or, worse, the company may be closed. Thus, on clinical grounds alone, information on exposures at work may be incomplete in some cases.

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Estimates of exposure to specific particulate contaminants have been reported from the measurements of the pulmonary retention of asbestos fibers (AF) (1–6), and have been proposed as a complement to the work history. The relative risk of mesothelioma in relation to lung content of asbestos fibers has been estimated in several studies (7–13).

A major problem in the interpretation of results of analysis of lung asbestos body and fiber content is the marked interlaboratory variation in body and fiber counts. There is no standard procedure when analyzing or reporting concentration between laboratories, and notable variations of lung concentration of fibers may occur between laboratories. Some studies will report all AF without any stratification on their length, and others will report with two levels of stratification, usually < 5 or ≥ 5 μm. Concentration data may be reported as arithmetic or geometric means. The analysis can be done by scanning electron microscopy or transmission electron microscopy, the latter instrument producing higher counts because of its higher resolution (14). Even when the same specimen is divided and analyzed by several different laboratories, markedly different results are obtained (15, 16).

Thus, although a wide variety of analytic results have been reported from different laboratories (10, 13–25), it is impossible to determine, without a properly selected reference group analyzed in the same laboratory, whether true elevations of fiber and body counts exist in a population of subjects with mesothelioma.

In a recent report (26) on work-related mesothelioma in Québec, we described 49 workers from the Québec chrysotile mines and 71 from other industries, either secondary (shipyard, insulation, factory, etc.) or tertiary (services such as plumber, electrician, etc.), and emphasized the increasing incidence of the disease. In this report we extend our previous observations in order to elucidate the types, quantities, and size distributions of asbestos fibers in the lung parenchyma of the subjects in the abovementioned study (71) and to compare them with a local reference population.

METHODS

Case Selection

All information related to lung tissue sample location, occupational histories, pathologic diagnoses, and other nominative information were available through the files of the Commission de la Santé et Sécurité du Travail du Québec (CSST Workers' Compensation Board). We wrote to pathologists at each hospital where the files of the CSST indicated that lung parenchyma and biopsy samples were likely to be held. Among the 120 workers compensated by the CSST (26) we obtained lung tissue specimens for 50. Forty-one specimens were lung parenchyma obtained at autopsy and nine were resections with both pleura and parenchyma. Another 10 additional workers were not included in the group of 50 because of an insufficient mass of lung parenchyma (mainly needle biopsies) for lung burden determination. There was no statistical difference (Mann-Whitney, $p > 0.1$) in the concentrations of asbestos fibers ≥ 5 μm between lung resections (11 samples) and lung parenchyma specimens (39 samples).

Among the 50 subjects with usable tissue included in this study, 12 worked in the chrysotile mining region of Asbestos Township, and data on lung asbestos concentration has been reported recently for this group (27). Eleven worked in the chrysotile mining region of Thetford Mines, and 27 worked in nonmining industries (26). The workers from other industries were mainly from Montréal, Québec, and Trois-Rivières-Sorel cities. Thirty-nine paraffin blocks and 11 wet tissue specimens of lung from the 50 workers were found in about 20 hospitals of the province of Québec. Most cases of mesothelioma from Asbestos Township were obtained from the Sherbrooke University Hospital Centre and from the Thetford Mines region from the Hospital Centre from the Région de l'Amiante. Five of the latter had wet fixed lung tissue available.

Lung fiber concentration of autopsy local reference population was estimated by analyzing 49 samples of wet lung tissue of either accidental death or death caused by acute myocardial infarction in men autopsied between 1990 and 1992 at Sherbrooke University Hospital Centre. According to medical records and autopsy reports, none of these men had known asbestos exposures. For those older than 45 yr of age we also looked in the Workers' Compensation Board files to ascertain that they were not compensated for any lung disease or that they were not cases submitted for compensation. These cases constituted the "autospy local reference population."

Certification of malignant mesothelial tumors was achieved by the pathologists from the hospitals where the tissues were obtained and validated by an expert pathologist retained by the CSST in the process of compensation. The clinical evolution of each case was consistent with that of mesothelioma.

The average age of the patients was 65.6 ± 10.5 yr. The duration of exposure was 26.4 ± 14.6 yr, and the time since last exposure was 14.9 ± 14.9 yr. Ten patients had less than 5 yr of exposure. The average age of the referent population was 49.4 ± 15.5 yr (range, 20 to 72 yr).

Specimen Preparation

The 99 paraffin blocks and wet tissues were analyzed for fibers by transmission electron microscopy (TEM). Specimens were prepared by a digestion filtration technique as previously described (27, 28).

Aliquots of 15 ml were filtered through a membrane filter 25-mm in diameter with a pore size of 0.45 μm (Millipore Corp., Bedford, MA). Filters were low-temperature ashed overnight, and the residue was resuspended in 50 ml distilled and deionized water. Suspensions were refiltered through a polycarbonate membrane filter 25-mm in diameter with a pore size of 0.2 μm (Nucleopore Corp., Pleasanton, CA). Filters were mounted on 200-mesh copper grids using a carbon replica technique (29).

For analysis of FB an aliquot of 25 ml was filtered through a cellulose membrane filter 25 mm in diameter with a pore size of 3 μm (Millipore®) using a Millipore filtration system.

Analysis

Grids were examined in a graphite sample holder mounted in a JEOL 100 CX transmission electron microscope equipped with a PGT System (PGT, NJ) IV® energy-dispersive X-ray spectrometer (EDS) for elemental analysis. They were observed in the transmission mode at a nominal screen magnification ×10,000 under an 80-kV accelerating voltage. Analysis of fibers was done in two steps. First, each visible fiber < 50 mm on the screen (1 mm = 0.09 μm) and having a length-to-diameter aspect ratio equal to or greater than 3:1 was recorded. Second, each fiber $\geq$ 50 mm on the screen was analyzed in a second step. Fiber types were identified by morphologic features and EDS spectrum. The length and diameter of all fibers were measured to the nearest 0.045 μm (0.5 mm on the screen) using two concentric circles (10 and 50 mm in diameter) drawn on the fluorescent screen. The number of grid openings scanned and the mass of lung tissue used was calculated to reach a limit of detection of 70 AF > 5 μm/mg of dry lung tissue.

The 3 μm membrane filter (for PCM analysis) was cleared and fixed on a glass slide using DMF (dimethyl formamide/acetic acid/water) and Eukitt (Kindler, Montreal, Canada) solutions. The FB were counted using a phase contrast microscope (Carl Zeiss) at a magnification ×312.

Statistical Analysis

The statistical analyses were performed with Minitab® statistical software (Minitab®, release 10). Geometric mean (GM) and geometric standard deviation (GSD) were used to describe the distributions of lung burden data in each group. Nondetected values were given half the limit of detection (35 AF/mg for asbestos fibers and 20 FB/g for ferruginous bodies) of the analytical method. Mood's nonparametric median test was used to compare median lung concentrations among the groups. The Mann-Whitney nonparametric median test was used to compare median lung concentrations between the lung resection and the lung parenchyma specimens. The 95% confidence interval (CI) was computed on the log-transformed data.

RESULTS

The geometric mean concentrations of fibers < 5 and $\geq$ 5 μm are shown, respectively, in Tables 1 and 2, and the GSD of all types of fibers, chrysotile, amosite, crocidolite, tremolite, all asbestos fibers, talc-anthophyllite, and other types of fiber (including cleavage fragments) of the three groups with mesothelioma and the reference population. For the latter group, Tables 1 and 2 also show the 95% CI concentrations.

As indicated in Tables 1 and 2, cases of mesothelioma from Asbestos Township had retained tremolite, chrysotile, crocidolite, and amosite fibers (27). Workers with mesothelioma cases from Thetford Mines retained only tremolite and chrysotile asbestos fibers. Workers with mesothelioma from other industries retained mainly amosite and crocidolite fibers. Very few amosite and crocidolite fibers were found in the reference group.

Concentrations of short and long chrysotile and tremolite fibers retained in the lungs of miners and millers were higher than in the lungs of workers from other industries. Median concentrations of all types of fiber in the lungs were statistically significantly different in the three groups of workers. However, median concentrations of FB were not statistically different, but these concentrations were statistically greater than those found in the reference population.

The GM concentrations and 95% CI limit of asbestos fibers of all sizes and the GM concentrations of the FB for all groups are shown in Table 3. The mean concentrations of AF < 5 and

TABLE 1

CONCENTRATION AND TYPE OF ASBESTOS FIBERS < 5 μm/mg DRY LUNG*

	Reference Population (*n* = 49)	Mesothelioma Cases: Other Industries (*n* = 27)	Asbestos Township (*n* = 12)	Thetford Mines (*n* = 11)	Probability Value
All Asbestos fibers	323 (6)	1,555 (6)	21,205 (3)	72,185 (15)	p < 0.001
95% CI	190–540				
Chrysotile	141 (5)	223 (5)	2,960 (6)	3,554 (15)	p = 0.001
95% CI	87–228				
Tremolite	144 (2)	104 (4)	8,103 (4)	57,641 (19)	p = 0.001
95% CI	87–240				
Crocidolite	40 (1)	162 (9)	1,224 (8)	35†	p < 0.001
95% CI	35–46				
Amosite	36 (1)	233 (6)	129 (5)	35†	p < 0.001
95% CI	35–38				
Talc or anthophyllite	117 (5)	158 (6)	866 (12)	3,692 (12)	p = 0.019
95% CI	76–180				
Other fibers	2,145 (6)	3,391 (3)	7,871 (2)	24,391 (4)	p < 0.001
95% CI	1,260–3,656				
Total fibers	2,981 (5)	6,568 (8)	36,098 (2)	146,532 (7)	p < 0.001
95% CI	1,823–4,876				

* Geometric Mean, shown in parentheses, = Geometric Standard Deviation; 95% CI = 95% Confidence Interval.
† No fibers detected.

≥ 5 μm in the workers with mesothelioma from all three groups were higher than the reference population.

The GM and GSD of diameters, lengths, and length to diameter (aspect) ratios of fibers < 5 μm and fibers ≥ 5 μm of each of the above-mentioned types of fibers are shown in Tables 4 and 5. The GM aspect ratios were in the following decreasing order for both cutoff lengths: chrysotile > crocidolite > amosite > tremolite > talc-anthophyllite ≈ other fibers. The log (base 10) of the aspect ratios of tremolitic fibers were compared with known minerals of tremolite such as a sample analyzed in a quality control program or samples that were from different locations (Figure 1). Length-to-diameter ratios of tremolite fibers extracted from lung parenchyma of workers from Asbestos Township and Thetford Mines were quite comparable to known asbestiform minerals of tremolite such as Californian tremolite and certainly larger than the cleavage fragments from the Nytal talc.

As an indication of which fiber type predominated for each subject, subjects were classified under the fiber type for which their lung concentration was highest. The distribution of subjects by this classification is shown in Table 6. For fibers < 5 μm, nonasbestos and tremolite fibers predominated. For fibers ≥ 5 μm, tremolite fibers predominated in the cases from Asbestos Township and Thetford Mines, whereas amosite and other types of fibers predominated in workers from other industries.

The frequency distribution of mesothelioma cases exceeding the 95% CI of the background population for each type of fiber < 5 and ≥ 5 μm is shown in Table 7. For instance, it can be seen in the second section of Table 7 that in Thetford Mines nine cases out of 11 exceed the reference population for tremolite asbestos and six out of 11 for chrysotile.

Individual concentrations of all fibers < 5 μm (third section in Table 7) were above the 95% CI of concentration of the reference population (i.e., > 546 AF/mg dry weight lung tissue) in 12 of 12 cases from Asbestos Township, in 11 of 11 cases from Thetford Mines, and in 18 of 27 cases from other industries. Individual concentrations of all fibers ≥ 5 μm (Table 7) were above

TABLE 2

CONCENTRATION AND TYPE OF ASBESTOS FIBERS > 5 μm/mg DRY LUNG*

	Reference Population (*n* = 49)	Cases of Mesothelioma: Other Industries (*n* = 27)	Asbestos Township (*n* = 12)	Thetford Mines (*n* = 11)	Probability Value
All asbestos fibers	103 (5)	612 (5)	5,541 (2)	11,294 (21)	p < 0.001
95% CI	66–161				
Chrysotile	56 (3)	69 (2)	508 (7)	353 (11)	p = 0.004
95% CI	40–79				
Tremolite	74 (4)	59 (2)	2,203 (4)	8,358 (19)	p < 0.001
95% CI	50–109				
Crocidolite	36 (1)	103 (6)	629 (4)	35†	p < 0.001
95% CI	35–37				
Amosite	37 (1)	226 (5)	107 (3)	35†	p = 0.003
95% CI	35–40				
Talc or anthophyllite	63 (2)	85 (3)	517 (6)	751 (8)	p < 0.001
95% CI	48–82				
Other fibers	272 (3)	145 (4)	654 (2)	949 (10)	p = 0.006
95% CI	186–398				
Total fibers	459 (4)	1,115 (4)	7,465 (2)	16,949 (10)	p < 0.001
95% CI	301–701				

* Geometric Mean, shown in parentheses, = Geometric Standard Deviation; 95% CI = 95% Confidence Interval.
† No fibers detected.

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TABLE 3

CONCENTRATION AND TYPE OF ASBESTOS FIBERS (ANY SIZE)/mg DRY LUNG*

	Reference Population (n = 49)	Cases of Mesothelioma: Other Industries (n = 27)	Asbestos Township (n = 12)	Thetford Mines (n = 11)
Asbestos fibers	507 (5)	2,497 (5)	27,342 (3)	89,680 (16)
95% CI	317–808	1,309–4,760	14,516–51,689	14,243–565,237
Ferruginous bodies	83 (6)	3,925 (14)	11,226 (7)	2,697 (37)
95% CI	49–142	1,404–10,971	3,381–37,235	270–26,903

* Geometric Mean, shown in parentheses, = Geometric Standard Deviation; 95% CI = 95% Confidence Interval. There was no overlap between 95% CI of reference population and 95% CI of each subset of cases of mesothelioma.

the 95% CI of concentration of the reference population (i.e., > 161 AF/mg dry weight lung tissue in the reference population) in 12 of 12 cases from Asbestos Township, in nine of 11 cases from Thetford Mines, and in 21 of 27 cases from other industries. Individual concentrations of FB (third section in Table 7) were above the 95% CI of the referent population ($\geq$ 140 FB/g) in 12 of 12 cases from Asbestos Township, in eight of 11 cases from Thetford Mines, and in 23 of 27 cases from other industries. The combination of either AF $\geq$ 5 μm or FB above referent cases was observed in 12 of 12 cases from Asbestos Township, in nine of 11 cases from Thetford Mines, and in 26 of 27 cases from other industries. Thus, overall, considering all cases of mesothelioma referred for possible workers' compensation, with known asbestos exposure, 92% of the cases had AF $\geq$ 5 μm or FB or both in excess of the 95% CI of the reference population of the same area and 96% of the cases of mesothelioma had AF (all sizes) or FB in excess of the reference population. The two workers who did not fall into the latter category had amosite and/or crocidolite concentrations greater than that of the reference population.

DISCUSSION

Because mesothelioma can develop years after relatively brief or relatively low exposure, analyses of the mineral fiber content of the lung are of interest in such cases. Some have suggested that fiber concentration in lung tissue, as analyzed by electron microscopy, is one of the best quantitative indicators of cumulated asbestos exposure (9, 30, 31). Others have emphasized the advantage of this method over measuring air concentrations or questionnaires because measurements of retained fibers provide information on the characteristics of the fibers that made up the exposure (10). It has been suggested that fiber analysis usually gives a more reliable estimate of past exposure to asbestos, even if a thorough work history is available (13); this is one of the points we wished to examine in this study. Finally, electron microscopic mineral fiber analysis is said to be of great value in detecting exposure to asbestos in cases where careful occupational histories are negative (32).

This issue is also of interest in the field of workers' compensation for occupational disease. For example, to consider accep-

TABLE 4

MORPHOLOGIC FEATURES OF FIBERS IN LUNG*

	Fibers < 5 μm: Number	Length GM (GSD)	Diameter GM (GSD)	Length/Diameter Ratio
Chrysotile				
Other industries	66	1.5 (1.8)	0.05 (1.6)	27 (2)
Asbestos Township	130	1.9 (1.6)	0.05 (1.4)	39 (2)
Thetford Mines	31	1.9 (1.7)	0.05 (1.5)	36 (2)
Tremolite				
Other industries	46	1.9 (1.7)	0.22 (2.2)	9 (2)‡
Asbestos Township	203	2.1 (1.6)	0.24 (2.0)	9 (2)
Thetford Mines	165	1.9 (1.6)	0.22 (2.1)	9 (2)
Crocidolite				
Other industries	80	1.8 (1.7)	0.10 (2.0)	18 (2)
Asbestos Township	70	2.1 (1.6)	0.10 (2.0)	21 (2)
Thetford Mines	0†			
Amosite				
Other industries	83	2.0 (1.7)	0.14 (2.0)	13 (2)
Asbestos Township	7	2.9 (1.6)	0.19 (2.0)	15 (2)
Thetford Mines	0†			
Talc or anthophyllite				
Other industries	75	2.0 (1.7)	0.43 (1.8)	5 (2)
Asbestos Township	69	2.0 (1.6)	0.30 (2.0)	6 (2)
Thetford Mines	34	2.1 (1.6)	0.34 (2.2)	6 (2)
Others				
Other industries	521	1.3 (1.7)	0.21 (2.1)	6 (2)
Asbestos Township	180	1.8 (1.6)	0.29 (2.0)	6 (2)
Thetford Mines	125	1.6 (1.6)	0.58 (2.2)	6 (2)

* Geometric Mean, shown in parentheses, = Geometric Standard Deviation.
† No fibers detected.
‡ $p < 0.001$ for Length/Diameter or tremolite versus other type of asbestos fibers.

TABLE 5

MORPHOLOGIC FEATURES OF FIBERS IN LUNG*

	Number	Fibers ≥ 5 µm Length GM (GSD)	Diameter GM (GSD)	Length/Diameter Ratio
Chrysotile				
Other industries	45	7.8 (1.5)	0.06 (1.9)	133 (2)
Asbestos Township	86	7.2 (1.5)	0.05 (1.7)	131 (2)
Thetford Mines	40	7.6 (1.4)	0.05 (1.3)	160 (2)
Tremolite				
Other industries	43	6.9 (1.5)	0.35 (2.2)	20 (2)‡
Asbestos Township	180	6.3 (1.4)	0.45 (2.0)	14 (2)
Thetford Mines	277	7.0 (1.5)	0.40 (2.1)	18 (2)
Crocidolite				
Other industries	107	7.7 (1.6)	0.11 (2.0)	71 (2)
Asbestos Township	101	6.8 (1.4)	0.13 (2.0)	50 (2)
Thetford Mines	0†			
Amosite				
Other industries	166	8.6 (1.7)	0.21 (1.9)	40 (2)
Asbestos Township	16	8.7 (2.1)	0.25 (1.8)	34 (2)
Thetford Mines	0†			
Talc or anthophyllite				
Other industries	56	7.0 (1.7)	0.97 (2.0)	7 (2)
Asbestos Township	71	7.0 (1.5)	0.57 (2.5)	12 (2)
Thetford Mines	39	7.3 (1.5)	0.93 (2.0)	8 (2)
Others				
Other industries	125	6.6 (1.3)	0.71 (2.4)	9 (3)
Asbestos Township	54	6.7 (1.4)	0.99 (1.9)	7 (2)
Thetford Mines	88	6.9 (1.4)	1.19 (2.0)	6 (2)

* Geometric Mean, shown in parentheses, = Geometric Standard Deviation.
† No fibers detected.
‡ $p < 0.001$ for Length/Diameter or tremolite versus other type of asbestos fibers.

tance of a mesothelioma claim by the Québec Workers' Compensation Board, one needs to show either a definite past exposure to asbestos at work or an excessive concentration of asbestos-related matter (AF and/or FB) in the lung tissue, indicative of a past exposure to asbestos. Given the low incidence of non-asbestos-related mesothelioma outside specific environmental exposures (32, 33), the established excess risk of mesothelioma in asbestos-exposed workers (5–13) and the increasing incidence of the disease in Québec workers (26) and other asbestos-exposed populations, the recognition of association in cases with long and well-documented exposure is not problematic. However, when exposures are relatively short, or forgotten by the patient because exposure occurred in the remote past (26), or exposures are conceivably not even known to the patient, it is desirable to have a set of criteria differentiating subjects with asbestos exposure and related disease from the reference population.

Such a set of criteria can be generated by analysis of AF and FB in lung tissue. Because there are marked interlaboratory variations in fiber counts, as noted above, it is crucial that a properly selected reference population analyzed by the same laboratory in exactly the same fashion be established to provide a baseline for comparison of cases and control subjects. Using a reference population selected from the Eastern Townships of Québec (an excellent comparison group for the Thetford Mines and Asbestos Township workers, and probably a reasonable comparison group for the other industry workers), we were able to demonstrate in this study that all 48 of 50 (96%) of our workers with mesothelioma had fiber concentrations greater than the upper 95% CI of the control group, and that in the remaining two cases the amosite and/or crocidolite concentrations exceeded the 95% CI of the control group. The latter findings almost always indicate an occupational exposure since these two amphiboles are pres-

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  N
100   -1-   Research Triangle Park (Bulk sample quality control)
100   -2-   Collection of A.D.
100   -3-   St-Lawrence
101   -4-   California
 33   -5-   Tremolite cleavage fragments (Nytal 300)
 50   -6-   Cleavage fragments of tremolite and talc fibres (Nytal)
203   -7-   Tremolite < 5 µm from Asbestos cases
180   -8-   Tremolite > 5 µm from Asbestos cases
 15   -9-   Tremolite < 5 µm from Thetford cases
  7   -10-  Tremolite > 5 µm from Thetford cases
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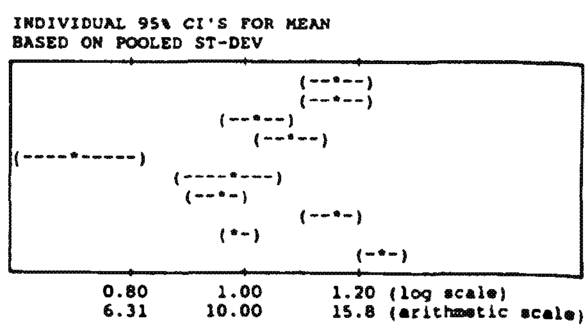

N = number of fibres

Figure 1. Log (base 10) of length-to-diameter ratio of tremolite fibers of different origins.

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TABLE 6

CASES OF MESOTHELIOMA

	Other Industries (*n* = 27)	Asbestos Township (*n* = 12)	Thetford Mines (*n* = 11)	Total (*n* = 50)
Frequency of highest concentration of fibers < 5 μm in lung				
Chrysotile	2*	3	0	5
Tremolite	0	6	8	14
Crocidolite	2	2*	0	4
Amosite	2	0	0	2
Talc anthophyllite	1	0	1	2
Other fibers	21*	3*	2	26
Frequency of highest concentration of fibers ≥ 5 μm in lung				
Chrysotile	2*	2	0	4
Tremolite	1*	7	10	18
Crocidolite	5*	2	0	7
Amosite	13*	0	0	13
Talc anthophyllite	5*	1	0	6
Other fibers	8*	0	1	9

* Subjects with equally high concentrations of two fiber types were classified in both.

ent in very low concentrations in the reference population. Thus, in our study all 50 workers with mesothelioma had analytic evidence of occupational asbestos exposure, confirming the occupational histories.

The one situation in which use of a reference population may fail is when exposure is to relatively pure chrysotile because chrysotile does not persist in lung tissue. Fortunately, pure chrysotile asbestos exposure appears to be rare, and most cases studied in the Québec miners and millers have substantial tremolite retention, reflecting common contamination of the chrysotile ore with the amphibole (1, 2, 6, 11, 21, 27).

An additional finding from our study is the marked difference in fiber types seen in the three mesothelioma groups. In the analysis done for this study at McGill, the concentration of tremolite was always greater than the concentration of chrysotile in the workers from Thetford Mines. These findings are similar to those reported by Churg and colleagues (34) who found, using different counting methods in a different laboratory, that a GM concentration of 180 × 10^6 (GSD, 4) fibers of tremolite (all sizes) and 34 × 10^6 (GSD, 4) fibers of chrysotile (all sizes) in 15 cases of mesothelioma from Thetford Mines. A greater concentration of tremolite than of chrysotile was previously reported by us in 10 of the 12 cases from Asbestos Township (26); however, these cases differ from the Thetford Mines cases by having a lower ratio of tremolite to chrysotile and also because of the presence of greater than background amosite and/or crocidolite in almost all the cases. In contrast, both this study and that of Churg and colleagues (34) have failed to find amosite or crocidolite in miners and millers from Thetford Mines. Despite the predominance of tremolite in the miners and millers, chrysotile levels exceeded those

TABLE 7

CASES OF MESOTHELIOMA

	Other Industries (*n* = 27)	Asbestos Township (*n* = 12)	Thetford Mines (*n* = 11)	Total (*n* = 50)
Frequency of cases exceeding the 95% CI of the background population for fibers < 5 μm in lung				
Chrysotile	12	11	9	32
Tremolite	7	12	11	30
Crocidolite	13	10	0	23
Amosite	16	5	0	21
Talc anthophyllite	10	9	9	28
Other fibers	13	11	10	34
Frequency of cases exceeding the 95% CI of the background population for fibers ≥ 5 μm in lung				
Chrysotile	10	9	6	25
Tremolite	2	12	9	23
Crocidolite	12	11	0	23
Amosite	20	6	0	26
Talc anthophyllite	7	8	8	23
Other Fibers	6	8	8	22
Number of cases exceeding 95% CI of reference population				
546 AF < 5 μm/mg	18	12	11	41
161 AF ≥ 5 μm/mg	10	12	9	31
808 AF all size/mg	19	12	10	41
142 FB/g	20	11	7	38
161 AF ≥ 55 μm/mg or 142 FB/g	25	12	9	46
808 AF any size/mg or 142 FB/g	25	12	11	48

Definition of abbreviations: AF = asbestos fibers; FB = ferruginous bodies.

in the reference population in 11 of 12 cases from Asbestos Township and nine of 11 cases from Thetford Mines. Lastly, the workers from the nonmining industries had relatively smaller amounts of chrysotile and tremolite (only four workers had chrysotile levels exceeding the background population) and much greater amounts of amosite and crocidolite, findings in accord with previous reports of cases of mesothelioma (7, 10, 11, 17). These data suggest that analyses of this type could potentially be used to classify sources of exposure with reasonable accuracy.

Data in the literature suggest that tremolite asbestos, and its nonasbestos analogues, have different biologic potential (35-37). Thus, it is important to distinguish among AF and cleavage fragments of tremolite. Fibers of tremolite extracted from lung parenchyma were compared with known asbestiform and nonasbestiform minerals, as shown in Figure 1. The log of the length-to-diameter ratios of tremolite fibers retained in the lungs of workers from Asbestos Township and Thetford Mines are comparable to acceptable reference tremolite asbestos fibers and had definitely longer length-to-diameter ratios than cleavage fragments of tremolite found in some talc samples, for instance. There is still a need to explain whether tremolite asbestos alone or actinolite asbestos or both are present in the mining settings. The distribution of the tremolite-actinolite asbestos fibers in the mines is still unclear.

Finally, there were no differences in concentrations of fibers between lung parenchyma obtained at autopsy and lung biopsies. Similar observations were reported by Tuomi (38) in one of his studies where he showed that with one exception results from autopsy and biopsy samples were equivalent.

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