American Industrial Hygiene Association Journal

ISSN: 0002-8894 (Print) (Online) Journal homepage: https://www.tandfonline.com/loi/aiha20

Evaluation of the Phase Contrast Microscopy Method for the Detection of Fibrous and Other Elongated Mineral Particulates by Comparison With a STEM Technique

J.G. SNYDER , R.L. VIRTA & J.M. SEGRETI

To cite this article: J.G. SNYDER , R.L. VIRTA & J.M. SEGRETI (1987) Evaluation of the Phase Contrast Microscopy Method for the Detection of Fibrous and Other Elongated Mineral Particulates by Comparison With a STEM Technique, American Industrial Hygiene Association Journal, 48:5, 471-477, DOI: 10.1080/15298668791385066

To link to this article: https://doi.org/10.1080/15298668791385066

Published online: 04 Jun 2010.

Am. Ind. Hyg. Assoc. J. 48(5):471-477 (1987)

Evaluation of the Phase Contrast Microscopy Method for the Detection of Fibrous and Other Elongated Mineral Particulates by Comparison With a STEM Technique

J.G. SNYDER, R.L. VIRTA and J.M. SEGRETI
U.S. Department of the Interior, Bureau of Mines, Avondale Research Center, 4900 Lasalle Rd., Avondale, MD 20782

The USPHS/NIOSH Membrane Filter Method is used to monitor for asbestos in occupational and mining atmospheres, and employs the phase-contrast optical microscope (PCM) that under optimum conditions has a resolution of approximately 0.25 μm. While amphibole cleavage fragments are usually visible by PCM, asbestos fibers (such as amosite and chrysotile) have finer widths that may render them invisible by PCM. In this study, personal air-monitoring filters containing chrysotile, amosite and amphibole cleavage fragments from various sources have been analyzed by PCM in accordance with the USPHS/NIOSH Method and scanning transmission electron microscopy (STEM) to assess the effectiveness of the PCM technique. Each STEM specimen was prepared using a direct-transfer technique to ensure that particle size distribution and concentration were not altered. STEM results for chrysotile samples are highly variable, with 9% to 81% of regulatory particles having widths smaller than 0.25 μm — the resolution of the optical microscope. Amosite samples have 27% to 38% of regulatory particles with widths below microscope resolution, indicating that routine particle counts by PCM on these samples would underestimate true fiber content by approximately one-third. All amphibole cleavage fragment samples had regulatory particles that would be observed by PCM. Multiplication factors have been suggested for application to routine counts by PCM to more accurately assess true particle content for mineral particulates on personal air-monitoring filters.

Introduction

To protect industrial and mine workers from excessive exposure to airborne asbestos, several versions of the Membrane Filter Method employing phase contrast optical microscopy (PCM) are in use for monitoring fiber concentration of asbestos in the air.[1-4] The U.S. Public Health Service/National Institute for Occupational Safety and Health (USPHS/NIOSH) method used in the United States involves pumping a specified volume of air through a membrane filter, treating the filter with a phthalate-oxylate solution to make it transparent, and counting the number of fibers within a determined area of the filter at a magnification of 450 to 500 X. Within the present guidelines set by the Mining Safety and Health Administration (MSHA) and the Occupational Safety and Health Administration (OSHA), asbestos dust exposure should not exceed two fibers per cubic centimeter of air [CFR Part 1910.1001, 30 CFR 55.1-1(b), 56.5-1(b), 57-5-1(b), and 71.202]. According to these guidelines, asbestos is defined as any particle that is 5 μm or greater in length; it has an aspect ratio (or length-to-width ratio) of 3:1 or greater and is one of six mineral types: chrysotile, amosite, crocidolite, tremolite asbestos, anthophyllite asbestos and actinolite asbestos.

Some nonasbestiform amphibole cleavage fragments fall within the fiber dimension criteria set by the MSHA and OSHA guidelines and are regulated erroneously as asbestos. Counting these nonasbestiform amphibole particles as "regulatory particles" is a limitation of MSHA and OSHA guidelines that employ the PCM technique. Another limitation is that some asbestiform amphibole and chrysotile fiber bundles and fibrils are below the resolution of the optical microscope. Because of those limitations, PCM fiber counts do not always represent the true number of fibers present. The ability of the PCM technique to detect mineral fibers is dependent on optical microscope resolution, fiber dimension criteria and width distribution. At 450 to 500 magnification, the resolution of the optical microscope is 0.2 μm to 0.4 μm, the actual value depending on the quality of the microscope. While larger particles and fiber bundles are resolved easily, chrysotile and amosite fibrils have mean widths of approximately 0.05 μm and 0.20 μ to 0.26 μm respectively.[5] To date, little has been done to quantify the number of fibers that may go undetected by the PCM technique during the routine analysis of occupational and mining personal air-monitor filters.

The purpose of this study is to evaluate the efficiency of the PCM technique for the detection of mineral particulates by comparing it to a technique employing the scanning transmission electron microscope (STEM), which has a typical resolution of less than 5 Å. This was accomplished by utilizing data on particle counts per square millimeter of filter area by both the PCM and STEM techniques and by analyzing particle dimension data obtained by STEM.

The Bureau of Mines undertook this study for the following reasons:

1) It is important to biomedical researchers to know the number and size distribution of the total particles present on personal air-monitoring filters. The term total particles refers to all particles with aspect ratio of 3:1 or greater with no length restriction, while the term regulatory particles refers to particles with aspect ratio of 3:1 or greater and lengths of 5 μm or greater. Specifically of interest is

TABLE I
Description of Personal Air-Monitor Filter Samples

Sample Designation[A]	Sample Source or Location	Mineralogy of Elongated Particles (3:1 aspect ratios)[B]
Chrysotile:		
1. C-O-1	Construction	100% chrysotile
2. C-O-2	Fiber packing	100% chrysotile
3. C-O-3	Cutting sheet asbestos	100% chrysotile
4. C-O-4	Cutting sheet asbestos	100% chrysotile
5. C-O-5	Mixing fiber for cement	92% chrysotile, 8% tremolite
6. C-M-1	Crushed stone operation mill	100% chrysotile
7. C-M-2	Crushed stone operation mill	100% chrysotile
8. C-M-3	Crushed stone operation open pit mine	100% chrysotile
9. C-M-4	Crushed stone operation	100% chrysotile
Amphibole asbestos:[C]		
10. AA-O-1	Vacuuming after asbestos removal	40% amosite, 35% chrysotile, 5% gypsum, 20% other
11. AA-O-2	Vacuuming after asbestos removal	40% amosite, 35% chrysotile, 5% gypsum, 20% other
12. AA-O-3	Insulation removal from ducting	35% amosite, 50% chrysotile, 15% other
13. AA-O-4	Insulation removal from ducting	60% amosite, 20% chrysotile, 4% gypsum, 16% other
Amphibole cleavage fragments:		
14. ACF-M-1	Underground mine, gold ore	100% cummingtonite
15. ACF-M-2	Open pit mine, iron ore	100% cummingtonite, hornblende, actinolite
16. ACF-M-3	Crushed stone operation	100% actinolite
17. ACF-M-4	Crushed stone operation	100% actinolite

[A]C = chrysotile; AA = amphibole asbestos (amosite); ACF = amphibole cleavage fragments (nonasbestiform amphiboles); O = occupational setting; M = mining setting.

[B]Qualitative chemistry determined on a particle-by-particle basis by STEM with EDXA.

[C]Amphibole asbestos samples; particle dimension data and particle count data in this report include only amphibole asbestos (amosite) particles determined by STEM/EDXA.

the comparison of total particles by TEM to regulatory particles measured by PCM, to gain insight on how many particles of undetermined hazard are missed using the present regulatory technique.

2) The number of regulatory particles visible in which the optical microscope is used depends on the particle type and width. Amphibole cleavage fragments tend to have large widths that allow particles to be visible by optical microscopy. Amosite and chrysotile fibers, on the other hand, have finer widths, some of which may not be observed by optical microscopy. Existing regulations make no corrections for the fact that these finer fibers are more likely to be underestimated during routine regulatory analysis in which the optical microscope is used. The number of fibers below the microscope resolution have been largely unknown for various occupational and mining atmospheres, as is the amount of variation among types of operations. In this study, STEM width data for regulatory particles of chrysotile, amosite and amphibole cleavage fragments from various sources have been analyzed to determine the percentage of these particles below optical microscope resolution. Multiplication factors have been determined from these percentages, as suggested by Rendall and Skikne,[6] to apply routine counts by PCM to help provide a more accurate estimation of regulatory particles present on air-monitoring filters.

3) Previous studies evaluating the PCM method have provided widely varying results, which may be due to differences in samples, specimen preparation techniques and methods of analysis. Winer and Cossette,[7] for example, showed that for various occupational settings 40 to 100 chrysotile fibers longer than 5 μm were observable by TEM for every one fiber counted by PCM; *i.e.*, approximately 1% to 2.5% of the chrysotile fibers were observed by PCM. Marconi *et al.*,[8] on the other hand, showed through TEM fiber characterization that approximately 90% of airborne chrysotile fibers generated from asbestos cement, insulation, and brake and clutch operations had diameters large enough to be visible by optical microscopy. Rendall and Skikne[6] indicate for amosite air-monitor filters from various occupational atmospheres that 30% to 50% of the fibers whose lengths and widths were measured by TEM had widths visible by PCM. The present study provides data on air-monitor filters containing chrysotile, amosite and amphibole cleavage fragments and utilizes a direct-transfer technique of specimen preparation to ensure that fiber distributions were not altered.

Samples

Seventeen personal air-monitor filters were obtained from OSHA and MSHA. The samples were characterized initially

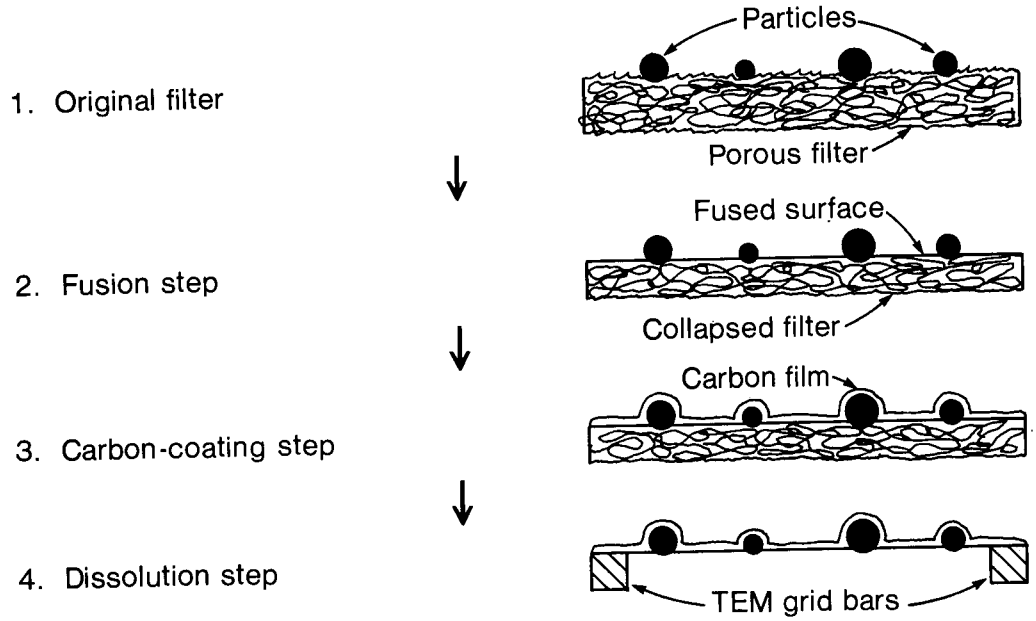

Figure 1 — Direct transfer of particles from Millipore filter to TEM grid in which the collapsed-membrane direct-transfer technique is used.

by STEM with the use of energy-dispersive X-ray analysis (EDXA) on a particle-by-particle basis. The samples described in Table I include airborne particulate samples of chrysotile from both occupational and mining sites, amphibole asbestos (amosite) from occupational sites and amphibole cleavage fragments from mining sites. Other elongated particles present in these samples include tremolite-actinolite, hornblende, gypsum and several unidentified phases.

Sample Preparation and Measurement

Phase Contrast Microscopy

Samples for PCM were prepared and analyzed according to the USPHS/NIOSH filter membrane method for evaluating airborne asbestos fibers.[1] Wedge-shaped sections were cut from air-monitor filters, mounted in a 1:1 dimethyl phthalate and diethyl oxylate solution on a microscope slide, and examined at 500 X with the use of PCM. Fields of view were selected in a grid pattern with the aid of a mechanical stage. The number of particles with an aspect ratio of 3:1 or greater for each field of view and length of 5 μm or greater was determined. This procedure was repeated until either 100 particles or 100 fields of view were encountered.

Electron Microscopy

The USPHS/NIOSH filter membrane method for evaluating airborne asbestos specifies the use of Millipore filters with 0.8 μm pore openings for sample collection. The rough surface of these filters makes direct electron microscopic examination impossible. Therefore, filter samples from both industrial and mining sites were prepared for STEM by using a direct-transfer method.[9] This method, which involves both a filter fusion and a dissolution step, allows transfer of particulate matter collected on Millipore filters to electron microscope grids. Wedge-shaped sections of filters are taped dust side up to glass slides. During the fusion step of this technique, the porous sponge-like structure of the Millipore filter was collapsed by exposure to acetone vapor in a petri dish for about 7 min. The fused filter wedges then were carbon coated in a vacuum evaporator to produce a continuous carbon film directly on the mineral particles and the fused membrane filters. Pieces of the carbon-coated filters were placed on TEM specimen grids, and the organic filter material was dissolved in a Jaffe washer in which acetone was used as a solvent. The result is a carbon film replica of the collapsed filter containing the mineral particles on the TEM grid. This technique is illustrated in Figure 1.

Particle losses in which this technique was used have been reported to not exceed 3%, usually.[10] There are still questions, however, as to whether the observed losses are particle-size dependent.[9,10] Smaller fibers reportedly would be able to penetrate into the filter membrane structure and be unavailable for entrapment in the carbon film after the filter was fused.

TEM specimen grid areas and particle lengths and widths were measured in the STEM mode. Measurements were made directly from the cathode ray tube over a magnification range of 1000 to 70 000 X. In cases where individual fibers or particles overlapped a grid square, the secondary electron mode was used for measurement of full particle length. The method of particle length and width measurement is adapted from Samudra *et al.*[11] When mats or bundles of asbestos whose aspect ratios exceeded 3 to 1 were encountered, they were measured as fibers to be consistent with the aspect ratio criteria used for the phase contrast microscope technique. At least 3 complete grid squares were examined for each sample, until 300 particles were encountered or 20 grid squares were examined.

Results and Discussion

For the purpose of general discussion, total particle size distributions from data determined by STEM analysis — including length, width and aspect ratio — are given in Figure 2. The data for the 17 individual samples are presented in four categories: chrysotile-occupational, chrysotile-mining, amosite-occupational and amphibole cleavage fragments-mining.

The length distributions for the four groups are similar, with the exception of the amosite-occupational category, which has a longer length distribution. All four groups have a significant percent of total particles that are less than 5 μm in length.

Width distributions for chrysotile-occupational and chrysotile-mining are similar, while width distributions for amosite-occupational and amphibole cleavage fragments-mining are slightly larger. This suggests that the chrysotile samples will have more total particles below a typical optical microscope resolution of 0.25 μm than will amosite-occupational and amphibole cleavage fragments-mining.

The aspect ratio distributions show the most difference between sample groups. The asbestos groups have more particles in the larger aspect ratio categories, while the amphibole cleavage fragments-mining group has few particles with aspect ratios greater than 10:1. These distributions are comparable to aspect ratio distributions observed in studies on crushed monominerallic samples of both asbestiform and nonasbestiform amphiboles and chrysotile.[12,13] The summary statistics for length, width and aspect ratio distributions are presented for individual samples in Table II.

The optimum analytical method for the detection of mineral particles present on air-monitor filters is one that can arrive at data closest to the true value of the number of fibers or particles present per square millimeter. The reliability of the PCM method presently used for monitoring industrial and mining atmospheres depends on microscope resolution, fiber width distribution and fiber criteria. The efficiency of the PCM method may be assessed by comparing it with a technique that optimizes the ability to observe fibers or microscope resolution. PCM has a typical working resolution of 0.25 μm. The STEM has been chosen for comparison purposes because of its resolution of less than 5 Å. The PCM technique outlined in the USPHS/NIOSH Filter Membrane Method suggests the measurement of particles longer than 5 μm with no minimum width restriction, although the effective width measurement limit is 0.25 μm; *i.e.*, the resolution limit of the optical microscope. Because there is no conclusive evidence to suggest that fibers with lengths less than 5 μm are biologically inactive,[14] the STEM method used in this study included counts of total particles with aspect ratios of 3:1 or greater with no length or minimum width restrictions.

Table III presents total particles detected by STEM and regulatory particles detected using PCM. The data are presented in particles per square millimeter of filter surface to allow direct comparison between the two data sets. Ratios of the data for these two methods of particle counting were determined to assess the efficiency of the PCM method.

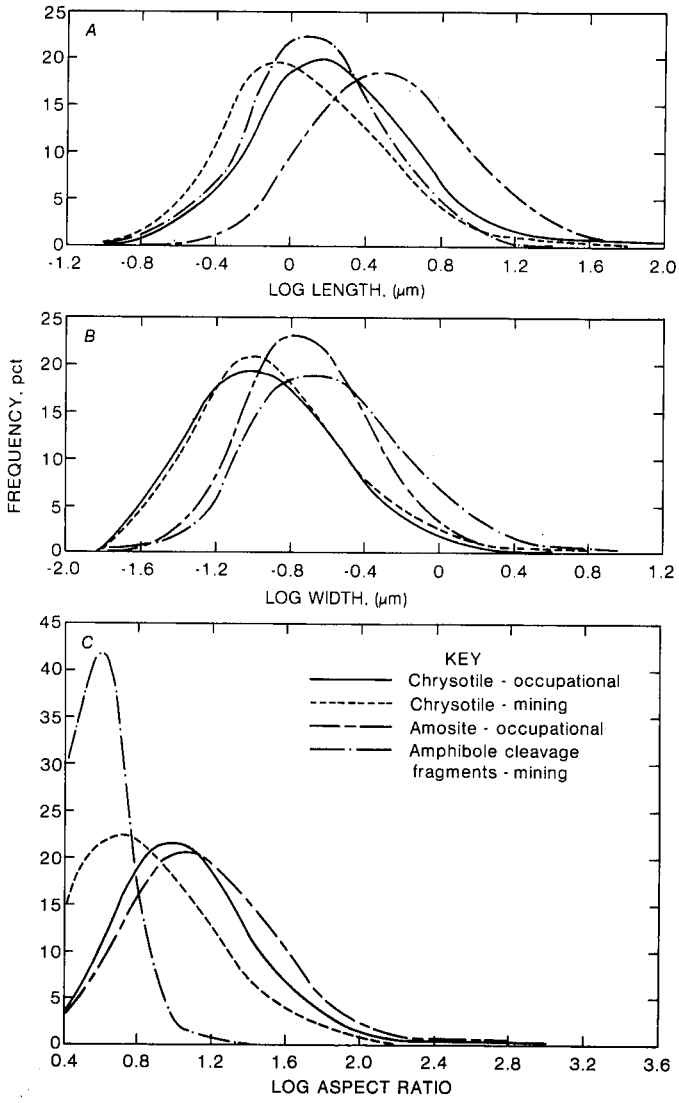

Figure 2 — Frequency distribution of log length, log width and log aspect ratio for the four occupational and mining categories. The number of particles in each category are as follows: chrysotile-occupational; n = 1562; chrysotile-mining; n = 1249; amosite-occupational; n = 1285; and amphibole cleavage fragments-mining; n = 1051.

Chrysotile occupational and mining site samples had the largest ratios and the largest variation in ratio, from 3:0 to 53:1. Except for amphibole cleavage fragment sample ACF-M-1, all of the amosite and amphibole cleavage fragment samples had ratios of less than 10:1. Sample ACF-M-1 had an unusually high ratio of 31.8:1, owing to the very small particle size, as indicated by the length (mean log length = 0.045) and width data (mean log = -0.698) shown in Table II. The chrysotile samples generally have larger ratios than the amphiboles. These results are reasonable because chrysotile fibers generally are narrower in width than amosite fibers and amphibole cleavage fragments. Hence more chrysotile would be below the resolution of the PCM. Chrysotile samples with low STEM-PCM ratios of particle counts were observed to contain fiber bundles and to have fewer individual fibrils than chrysotile samples with larger ratios.

TABLE II
Summary Statistics for Length, Width and Aspect Ratio Distributions for 17 Air-Monitor Filter Samples From Various Sources

Sample Designation	Log Length (μm)			Log Width (μm)			Log Aspect Ratio		
	Mean	Min	Max	Mean	Min	Max	Mean	Min	Max
Chrysotile:									
1. C-O-1	0.031	-0.775	1.372	-0.911	-1.553	0.663	0.942	0.047	1.823
2. C-O-2	0.184	-0.903	1.796	-0.886	-1.505	0.706	1.070	0.477	2.394
3. C-O-3	0.483	-0.425	1.879	-0.668	-1.618	0.487	1.151	0.484	2.174
4. C-O-4	0.061	-0.723	1.365	-0.706	-1.598	0.888	0.767	0.198	1.869
5. C-O-5	0.380	-0.557	1.487	-0.619	-1.461	0.310	0.999	0.439	2.045
6. C-M-1	0.267	-0.648	1.247	-0.950	-1.493	-0.230	1.217	0.046	2.206
7. C-M-2	0.295	-0.720	2.000	-0.861	-1.799	0.275	1.157	0.118	3.799
8. C-M-3	0.597	-0.609	2.173	-0.630	-1.524	0.468	1.227	0.556	3.072
9. C-M-4	0.153	-0.861	1.668	-1.120	-1.794	0.606	1.273	0.477	2.528
Amphibole asbestos:									
10. AA-O-1	0.610	-0.522	1.996	-0.654	-1.440	0.285	1.264	0.553	2.816
11. AA-O-2	0.715	-0.456	2.098	-0.638	-1.516	0.143	1.353	0.514	2.286
12. AA-O-3	0.666	-0.388	1.931	-0.588	-1.786	0.305	1.255	0.254	2.620
13. AA-O-4	0.647	-0.255	1.794	-0.595	-1.416	0.125	1.242	0.301	2.393
Amphibole cleavage fragments:									
14. ACF-M-1	0.045	-0.731	0.986	-0.698	-1.516	0.264	0.743	0.445	1.731
15. ACF-M-2	0.369	-0.420	1.620	-0.359	-1.237	1.118	0.728	0.477	1.612
16. ACF-M-3	0.339	-0.691	1.413	-0.355	-1.469	0.852	0.695	0.025	1.506
17. ACF-M-4	0.214	-0.411	0.929	-0.476	-1.452	0.149	0.690	0.477	1.230

The relatively low STEM/PCM ratios and the small variation between ratios for amosite samples from occupational atmospheres and amphibole cleavage fragment samples from mining atmospheres indicate that PCM is considerably more effective for detecting amosite and amphibole cleavage fragments than chrysotile. This is reasonable because of the larger particle widths of amosite fibers and amphibole cleavage fragments. It should be mentioned again, however, that the PCM technique does not provide a means of discrimination between amosite and cleavage fragments.[15]

One consideration in comparing these PCM and STEM data is the effect of sample preparation on the STEM analy-

TABLE III
Total Particles With Aspect Ratios of 3:1 or Greater per Square Millimeter Detected by STEM *vs.* Regulatory Particles per Square Millimeter Detected by PCM

Sample Designation	A Total Particles (particles per square millimeter) STEM	B Regulatory Particles (particles per square millimeter) PCM	A/B
Chrysotile:			
1. C-O-1	9814	505	19.4
2. C-O-2	14 808	563	26.3
3. C-O-3	1425	474	3.0
4. C-O-4	9392	222	42.3
5. C-O-5	7352	497	14.8
6. C-M-1	6251	117	53.1
7. C-M-2	1941	127	15.3
8. C-M-3	3431	416	8.3
9. C-M-4	4045	80	50.6
Amphibole asbestos:			
10. AA-O-1	3299	642	5.1
11. AA-O-2	4263	990	4.3
12. AA-O-3	1838	471	3.9
13. AA-O-4	5071	1077	4.7
Amphibole cleavage fragments:			
14. ACF-M-1	1590	50	31.8
15. ACF-M-2	1331	196	6.8
16. ACF-M-3	1489	187	8.0
17. ACF-M-4	597	74	8.1

sis. The direct-transfer method used in this study should not have altered the fiber size distribution, and particle losses should have been minimal.[9] Techniques that employ ultrasonic vibration, however, may break up and disperse particle bundles. This has been shown to be true particularly for chrysotile, because the bundles are more likely to disrupt lengthwise upon ultrasonic treatment than amphibole asbestos.[16,17] Chrysotile air-monitor samples examined in this study contain fiber bundles, composed of many individual fibrils. Preliminary experiments at the Bureau of Mines show that these fibrils are released during ultrasonic treatment. If ultrasonic treatment had been used during specimen preparation of these samples, the STEM/PCM ratios might have been several orders of magnitude larger.

As mentioned previously, those regulatory particles with widths less than 0.25 μm will be below the resolution of the PCM; these fibers will influence the effectiveness of the PCM technique. Rendall and Skikne[6] have suggested that the effectiveness of the PCM method may be increased by the application of multiplication factors to the routine counts of asbestos as defined:

$$\text{multiplication factor, } F = \frac{A + B}{B} \qquad (1)$$

where A = particles with lengths $\geq$5 μm and widths below the resolution of the optical microscope (submicroscopic);

B = particles with lengths $\geq$5 μm and widths above the resolution of the optical microscope (microscopic).

In Equation 1, the minimum length of 5 μm used in this calculation corresponds to the lower limit of the PCM counting criteria for asbestos. Rendall and Skikne[6] suggest that since it is not practical to perform TEM analysis on every sample on a routine basis because of the time, expertise and expense involved, multiplication factors may be applied to routine counts by PCM to help provide a more accurate estimation of regulatory particles present on air-monitor filters. They suggest that, because of size distribution variations of fibers from different operations and mineral types present, multiplication factors would need to be derived for each specific area or operation in which TEM is used. Multiplication factors are given in Table IV for each air-monitor sample. These values supply information on the effectiveness of PCM and are not necessarily for use in routine PCM analysis. Optical microscope resolution used in the calculation of the multiplication factors was assumed to be 0.25 μm.

Multiplication factors for chrysotile air-monitor samples from both occupational and mining sources were close to 1.5, with the exception of samples C-O-1 and C-O-3 with higher values of 2.7 and 5.3, respectively. Both of these samples are from industrial sources and contain fewer fiber bundles than other chrysotile samples. Amosite samples from occupational sources have multiplication factors that also are close to 1.5, indicating approximately one-third of the regulatory particles are below the resolution of the optical microscope. All amphibole cleavage fragment samples from mining sources have multiplication factors of 1, indicating that they contain no regulatory particles below the microscope resolution of 0.25 μm.

These data show that although there are some variations in the derived multiplication factors, general trends do exist. Our data are in good agreement with those of Rendall and Skikne,[6] although they assume a microscope resolution of

TABLE IV
Evaluation of PCM Method for the Detection of Regulatory Particles from STEM Particle Dimension Data[A]

Sample Designation	A % Particles Below Width of 0.25 μm	B % Particles Above Width of 0.25 μm	Multiplication Factor (A+B)/B
Chrysotile:			
1. C-O-1	63	37	2.7
2. C-O-2	32	68	1.5
3. C-O-3	30	70	1.4
4. C-O-4	81	19	5.3
5. C-O-5	21	79	1.3
6. C-M-1	9	91	1.1
7. C-M-2	41	59	1.7
8. C-M-3	30	70	1.4
9. C-M-4	11	89	1.1
Amphibole asbestos:			
10. AA-O-1	38	62	1.6
11. AA-O-2	35	65	1.5
12. AA-O-3	27	73	1.4
13. AA-O-4	37	63	1.6
Amphibole cleavage fragments:			
14. ACF-M-1	0	100	1.0
15. ACF-M-2	0	100	1.0
16. ACF-M-3	0	100	1.0
17. ACF-M-4	0	100	1.0

[A]Regulatory particles are those with aspect ratios of 3:1 or greater, and lengths of 5 μm or greater.

0.4 μm. In their study, a chrysotile mine air-monitor filter sample resulted in a factor of 4.3, and four amosite mine samples ranged from 1.9 to 3.0. Mixed-fiber-type samples included one from an insulation factory with a factor of 4.4, and lung tissue from a mine worker with 2.7.

Although it may be impractical to determine these factors accurately for individual operations, it is plausible to determine general factors for types of operations. Clearly, multiplication factors of 1 are applicable to the amphibole cleavage fragment samples from mining sources. General multiplication factors of 1.5 were determined for amosite and chrysotile samples from occupational sources and chrysotile samples from mining sources. Chrysotile samples C-O-1 and C-O-3 from occupational atmospheres yielded higher multiplication factors of 2.7 and 5.3, respectively. While these values are not extremely larger than values for the other asbestos samples, they indicate that variations in fiber widths distributions of chrysotile samples do occur. Unless multiplication factors are known for specific sites, regulation of these types of samples should include analysis by STEM or TEM.

Summary and Conclusion

In the analysis of total fibers (particles with $\geq$3:1 aspect ratio), PCM alone does not detect all mineral fibers adequately; STEM/PCM particle count ratios indicate that a significant number of particles were missed in each sample. This is particularly true for samples containing chrysotile, which had STEM/PCM ratios of up to 53:1. Most samples containing amosite or amphibole cleavage fragments had ratios of less than 10:1.

To assess the efficiency of PCM for the detection of regulatory particles on personal air-monitor filters, general multiplication factors were determined for the four sample types. Multiplication factors of 1 for amphibole cleavage fragment samples from mining sites indicated that the PCM technique is adequate because no regulatory particles were below the resolution of the optical microscope. Multiplication factors of 1.5 were determined for amosite and chrysotile samples from occupational sites and chrysotile samples from mining sites. This indicates that approximately one-third of the regulatory particles in these samples were below the resolution of the optical microscope.

Exceptions arise when air-monitor samples contain many thin particles, as was the case with some chrysotile samples in this study. In addition, many chrysotile particles observed by PCM were determined to be comprised for hundreds of individual fibrils when observed by STEM or TEM. For these reasons — unless multiplication factors are known for specific sites — regulation of chrysotile should include analysis by STEM or TEM. Sample preparation should include a direct-transfer technique that does not alter the original particle concentration and size distribution.

References

1. **U.S. Department of Health, Education and Welfare/Public Health Service/Center for Disease Control/National Institute for Occupational Safety and Health:** *USPHS/NIOSH Membrane Filter Method for Evaluating Airborne Asbestos Fibers* by N.A. Leidel, S.G. Bayer, R.D. Zumwalde and K.A. Busch. (DHEW/NIOSH Technical Report No. 79-127). Cincinnati, Ohio: NIOSH, 1979.
2. **Ontario Ministry of Labour:** *Regulation Respecting Asbestos* [created under the Occupational Health and Safety Act, Revised Statutes of Ontario, 1980, Chapter 321, filed 20 August 1982]. Toronto, Ontario Ministry of Labour: Occupational Health and Safety Division, 1982.
3. **Asbestos International Association:** *Reference Method for the Determination of Airborne Asbestos Fibre Concentrations at Workplaces by Light Microscopy (Membrane Filter Method)* [AIA Health and Safety Publication, Recommended Technical Method No. 1 (RTM1)]. London: AIA, 1979.
4. **International Organization for Standardization:** *Determination of Airborne Inorganic Fibre Concentrations in Workplaces by Light Microscopy (Membrane Filter Method)* edited by U. Teichert. [Draft International Standard ISO/DIS, ISO/TC146/SC2/W65]. Neuss 1, West Germany: Asbestos Institute for Occupational and Environmental Safety and Health, 1981.
5. **Gibbs, G.W. and C.Y. Hwang:** Dimensions of Airborne Asbestos Fibers. In *Biological Effects of Mineral Fibers* edited by J.C. Wagner. *IARC Sci. Pub. 30*:69-78 (1980). [Switzerland].
6. **Rendall, R.E.G. and M.I. Skikne:** Submicroscopic Fibres in Industrial Atmospheres. In *Biological Effects of Mineral Fibers,* Vol. II, edited by J.C. Wagner. *IARC Sci. Pub. No. 30*:837-843 (1980). [Switzerland].
7. **Winer, A.A. and M. Cossette:** The Effect of Aspect Ratio on Fiber Counts: A Preliminary Study. *Ann. N.Y. Acad. Sci. 330*:661-672 (1979).
8. **Marconi, A., E. Menichini and L. Paoletti:** A Comparison of Light Microscopy and Transmission Electron Microscopy Results in the Evaluation of the Occupational Exposure to Airborne Chrysotile Fibres. *Ann. Occup. Hyg. 28(3)*:321-331 (1984).
9. **Ortiz, L.W. and B.L. Isom:** Transfer Technique for Electron Microscopy of Membrane Filter Samples. *Am. Ind. Hyg. Assoc. J. 35(7)*:423-425 (1974).
10. **Chatfield, E.J.:** *Measurement of Asbestos Fibre Concentrations in Ambient Atmospheres.* Royal Commission on Asbestos Study No. 10, Ontario, Canada, 1983. [ISBN 0-7743-8175-2].
11. **Samundra, A.V., C.F. Harwood and J.D. Stockham:** *Electron Microscope Measurement of Airborne Asbestos Concentrations: A Provisional Methodology Manual* (EPA 600/2-77-178). Research Triangle Park, N.C.: USEPA, 1977.
12. **Campbell, W.J., R.L. Blake, L.L. Brown, E.E. Cather and J.J. Sjoberg:** Selected Silicate Minerals and Their Asbestiform Varieties [U.S. BuMines Inf. Cir. 8751]. Washington, D.C.: Bureau of Mines, 1977.
13. **Siegrist, H.G., Jr. and A.G. Wylie:** Characterizing and Discriminating the Shape of Asbestos Particles. *Environ. Res. 23*:348-361 (1980).
14. **Pott, F.:** Animal Experiments in Biological Effects of Mineral Fibers, In *Biological Effects of Mineral Fibers* edited by J.C. Wagner. *IARC Sci. Pub. 30*:261-272 (1980). [Switzerland].
15. **Wylie, A.G., R.L. Virta and E. Russeck:** Characterizing and Discriminating of Airborne Amphibole Cleavage Fragments and Amosite Fibers: Implications for the NIOSH Method. *Am. Ind. Hyg. Assoc. J. 46(4)*:197-201 (1985).
16. **Spurny, K.R., W. Stober, H. Opiela and G. Weiss:** On the Problem of Milling and Ultrasonic Treatment of Asbestos and Glass Fibers in Biological and Analytical Applications. *Am. Ind. Hyg. Assoc. J. 41*:198-203 (1980).
17. **Spurny, K.R., H. Opiela and G. Weiss:** On the Milling and Ultrasonic Treatment of Fibres for Biological and Analytical Applications. In Biological Effects of Mineral Fibers edited by J.C. Wagner. *IARC Sci. Pub. 30*:931-933 (1980). [Switzerland].

19 September 1985; Revised 24 October 1986