25 September 1972



INDUSTRIAL HYGIENE DIVISION
PROCEDURE NO. L-27

Title: Collecting and Counting Airborne Asbestos Fibers

References:
(a) NAVSHIPS INST 5100.26, Subj: Asbestos Exposure Hazards; Control of, of 9 Feb 1971
(b) Documentation of the Threshold Limit Values; American Conference of Governmental Industrial Hygienists; 3rd Edition 1971
(c) Edwards, G. H., and Lynch, J. R., - The Method used by the U.S. Public Health Service for Enumeration of Asbestos Dust on Membrane Filters. Annals of Occupational Hygiene 11:1-6 (1968)
(d) Criteria for a recommended standard for Occupational Exposure to Asbestos - HSM 72-10267

Discussion: In order to prevent the occurrence of occupational lung diseases (asbestosis, malignancies) among asbestos workers, a more accurate method for evaluating exposure to asbestos fibers in air than that provided by impinger sampling and light field dust counting is required.

The method describes a currently accepted procedure for the collection of airborne asbestos fibers on membrane filters and counting of the fibers by phase contrast microscopy.

Equipment:

A. Sampling:

1. Vacuum Pump, Bell and Gossett Duraire 1/10 HP, 110V, Model No. V200.

2. Staplex, Unico 110V, Model No. 30.

3. Monitaire Sampler, Mine Safety Appliance Co., Pittsburgh, Pa.

4. Membrane filters, millipore Corporation, Bedford, Massachusetts.

   a. 37mm white, cellulose ester, 0.8μ pore size, AAWP 03700, mounted in plastic field monitor MAWP 037A0.

   b. 47mm, cellulose ester for metal open face filter holder (Millipore Corporation Catalog #AAWP 04700) 0.8μ pore size.

5. Filter holder, aerosol type, open face, 47mm, Millipore Corporation, Catalog #XX50-047-10.

6. Plastic petri dishes for 47mm filters.

7. Forceps, w/o serrated tips.

8. Scalpel or surgical scissors.

9. Stop watch.

B. Slide Preparation:

1. Microscope slides, precleaned, preferably with frosted end, 1" X 3" (25 X 75mm).

2. Cover slips, No. 1-1/2, 24 X 30mm square, 0.16 - 0.19mm thick.

3. Acetone.

4. Diethyl oxalate.

5. Dimethyl phthalate.

6. Immersion oil bottle with glass rod fixed to the cap.

C. Phase Microscopy:

1. AO Microstar microscope, complete with binocular eyepiece, AO #1177 10X oculars, (Huygenian) GSA Catalog #GS-005-07872, 10X bright field objective, 45X phase contrast objective, 100X oil immersion objective, bult-in illuminator with transformer, phase condenser, phase annulus and phast aperture viewing unit. A green Tiffan filter (A. O. Catalog #408) is installed over the illuminator lens.

2. Porton reticle installed above the lens of the ocular. Calibrated field area 0.0056MM^2.

3. Stage micrometer, 0.1mm and 0.01mm calibration lines or equal.

Procedure:

A. Sample Collection:

1. A blank millipore filter must be reserved from each set of filters taken to th field. 37mm filters are carried in their filter holders while 47mm filters are kept in the packing container until needed.

2. Pump and filter system should be calibrated in the laboratory before and after field use. Laboratory testing should be with a 1-liter burette or wet-test meter.

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3. During field sampling airflows should be checked before and after each filter sample. Attach the millipore membrane filter to the sampling pump and adjust the air flow of each pump with a calibrated rotameter. The 37mm filters are used for personnel sampling at a sample rate of 1 to 4 liters per minute. The 47mm filter is used for general sampling at a flow rate of 20 to 35 liters per minute.

4. Sampling time will be dependent on the expected airborne asbestos fiber concentration. However, samples should be collected for a minimum of 15 minutes and a maximum of 480 minutes.

5. Mark the filter holder with code number and date as follows:

   Capital letter for operation, i.e., F for fabrication; I for installation; R for rip-out and month and day separated by a slash and a capital letter for each sample, viz: R 10/5 A. The 37mm filter holder is used in open-face mode, i.e., with the top cover removed, with filter inclined downward.

6. After sampling is concluded replace the top cover of the 37mm filter holder and place the 47mm filter in a petri dish and fasten sides with tape.

7. Record all sampling data, including operation, flow rate, sample time, materials used in operation and pump used and environmental controls in effect on the survey report form.

8. Transport the filters back to the laboratory for evaluation, keeping the filters face up.

B. Sample Preparation:

1. Prepare a mounting medium of 1:1 dimethyl phthalate and diethyl oxalate in an oil bottle with a glass rod fixed to the cap.

2. Add approximately 5% by weight of crushed white millipore filter material and stir to dissolve. The amount of filter material added may be varied to produce a desired viscosity. Final solution should be clear.

3. Clean the slide and cover glass with acetone.

4. Put a drop of mounting medium on the center of the slide and spread it with the glass rod. Use only enough fluid to make the filter section transparent and yet stick to the cover glass.

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5. Cut a pie-shaped wedge or sector from the filter approximately 10% of filter area) and place it on the sampling medium, sample side up. The filter may be cut with a clean scalpel or small surgical scissors.

6. Place the cover glass on the filter and press slightly in the center with the end of the tweezers. This will force any bubbles to the edges and distribute the mounting medium to uniform thickness before the filter dissolves. It also ensures wetting of the entire filter wedge. Mark the outline of the wedge on the cover slip with a felt-tip pen.

7. Allow the slide to stand until the filter wedge clears.

8. Sample should be counted within two days of mounting.

C. Phase Contrast: (See attached procedure on page 5)

D. Sample Counting:

1. Count all fibers greater than 5 microns in length. Any particle having an aspect ratio of three or greater is considered a fiber.

2. Count the fibers extending outside the edges of the counting area on any two sides to average the count. Do not count the fibers extending outside the edges of the counting area on the remaining two sides.

3. As many fields as required to yield at least 100 fibers should be counted. In general, the minimum number of fields should be 20 and the maximum 100. Do not count any field containing over 20 fibers.

4. Calculate the airborne concentration as indicated on the attached sample evaluation sheet (page 6) and transfer data to the survey sheet.

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PROCEDURE FOR ALIGNING MICROSCOPE AND PHASE CONTRAST ASSEMBLY

1. Switch on illuminator and set to 5.5 volts.

2. Install phase annulus in bright field mode (pulled out).

3. Place specimen slide on stage and focus with 10X objective.

4. Align condenser

   a. Remove either eyepiece.

   b. Look down eye tube (from a "distance"), to see if iris leaves are centered (leaves at outer edge of field). If not centered, use centering screws on condenser to center. When centered, open iris leaves to show at periphery.

5. Align illuminator

   a. Adjust focus of condenser by knob to focus on field diaphram iris leaves.

   b. Center iris leaves using centering screws on illuminator.

   c. When centered, open leaves until they just leave the field of view.

6. Repeat using 45X to critically adjust to that objective.

   Microscope is now aligned.

7. Align phase

   a. Insert phase annulus in phase mode (pushed in).

   b. Swing in phase aperture viewing unit and focus.

   c. Center light ring inside dark ring using centering wrenches. (Pull straight out on removal).

   d. Swing viewer out of field.

8. Phase alignment should be checked occasionally depending on use.

9. Start counting.

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COLLECTION AND COUNTING AIRBORNE ASBESTOS FIBERS (PHASE CONTRAST MICROSCOPY METHOD)

Airborne Asbestos Fiber Concentration Data Sheet for SAMPLE #: _____

Sample Location:

Operation:

Date Collected: _____ Filter Size _____ mm. Type Air Sampler: _____ Serial # _____

Air Sample Flow Rate: _____ liters/min. Time: _____ min. Total Volume _____ liters

FIBER COUNT:
(> 5 μm in length)

Field #			
	1		
	2		
	3		
	4		
	5		
	6		
	7		
	8		
	9		
	10		
	11		
	12		
	13		
	14		
	15		
	16		
	17		
	18		
	19		
	20		
Total			
Average			
Blank			
Net			

FORMULA:

$$\text{Airborne Asbestos (fibers/ml)} = \frac{(\text{net fiber count})\ (\text{effective filter area, } mm^2)}{(\text{Microscope field area, } mm^2)\ (\text{Tot. air sample Volume, L})\ (1000\ ml/L)}$$

CALCULATIONS:

Effective filter area
37 mm filter = 855 mm^2
47 mm filter = 960 mm^2

RESULTS:

Airborne asbestos concentration = _____ fibers/ml.air.

Collected by: _____
Analyzed by: _____
Date: _____

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ASBESTOS FIBER COUNTING RING

