ENVIRONMENTAL LABORATORY APPROVAL PROGRAM
CERTIFICATION MANUAL

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Polarized-Light Microscope Method for Identifying and Quantitating Asbestos in Surfacing Material Containing Vermiculite Bulk Samples	05/06/16	1 of 42	198.8

1. **Introduction.** This method is for the analysis of asbestos in **Surfacing Material** as defined in 12 NYCRR Part 56 (NYS Industrial Code Rule 56) such as Sprayed-On Fireproofing Containing Vermiculite (SOF- V) by Polarized-Light Microscopy (PLM).

1.1. **Background**. Prior to 1990, the majority of vermiculite used in building materials, including SOF, originated from a mine located in Libby, Montana, U.S.A (Meeker, 2003, EPA 2001). Depending on the date of production, the vermiculite shipped from the Libby mine may have contained various concentrations of amphibole minerals, including tremolite and actinolite, which are regulated by the Environmental Protection Agency (EPA) under the 1987 AHERA definition of 'Asbestos'. In addition, this vermiculite contains richterite and winchite, which are unregulated amphiboles under the AHERA definition for asbestos (AHERA, 1987, Meeker, 2003). It has long been recognized that vermiculite particles interfere with the accurate identification and quantitation of asbestos fibers during sample analysis, which increases the likelihood of reporting of false negative results for asbestos in samples of SOF-V and other vermiculite containing materials (EPA, 2012). These uncertainties, combined with the potential public health concern (EPA, 2009) arising from erroneous analytical interpretation, led to the development and validation of this ELAP method, Item 198.8. The method is based on a draft document prepared in 2000 under contract with the EPA (Chatfield, 2000). It utilizes validated techniques to remove vermiculite and other SOF-V constituents prior to analysis, thereby improving the statistical confidence of the analytical result at low concentrations of asbestos. At this time, NYS DOH's Item 198.8 and RJ Lee Group's Method 055 are the only methods approved for the analysis of vermiculite-containing materials for asbestos. Laboratories that would like to perform analysis for the detection and quantitation of asbestos content in surfacing material must seek accreditation for Item 198.8. Item 198.8 will be validated for use on other materials that contain vermiculite in the future (Table 1).

1.2. **Objective**. Item 198.8 incorporates a two-step approach for the identification and quantitation of chrysotile and amphibole asbestos, including Libby amphiboles, in surfacing material containing vermiculite (SM-V). The first step utilizes gravimetric reduction including ashing to remove the organic materials and dilute acid treatment to remove gypsum and cement from SM-V. The residue is then examined by PLM for the presence of chrysotile, which is quantitated by point counting. If the concentration of chrysotile is found to exceed 1%, the material is considered asbestos containing material (ACM) and the analysis is terminated. If chrysotile is either not detected, or is found at a concentration less than 1%, then the analysis is continued to determine the concentration of amphibole asbestos. Heavy liquid centrifugation is used to separate particles with densities exceeding 2.75 g/cc from the majority of the less dense matrix components. This results in a centrifugate that contains any amphibole that was present in the original sample. The concentration of the amphibole can then be reliably determined by PLM and point counting. The total asbestos content is obtained by adding the concentration of asbestos quantitated in step one (chrysotile) with the asbestos quantitated in step two (amphibole). If the final

concentration is determined to be greater than 1%, the material is designated as ACM.

1.3. **Definitions**.

1.3.1. **Amphibole Asbestos**. Asbestiform varieties of amphibole including, but not limited to, amosite (cummingtonite-grunerite), crocidolite (riebeckite), anthophyllite, tremolite, actinolite, richterite and winchite (IMA, 1978 and 1997).

1.3.2. **Asbestiform**. Specific type of mineral fibrosity in which the fibers and fibrils possess high tensile strength and flexibility (ISO 10312, 1995).

1.3.3. **Asbestos**. "Asbestos" refers to the asbestiform varieties of: chrysotile (serpentine); crocidolite (riebeckite); amosite (cummingtonite-grunerite); anthophyllite; tremolite; and actinolite (AHERA, 1987).

1.3.4. **Asbestos-Containing Materials**. "Asbestos-containing materials" (ACM) means any material or product that contains more than 1 percent asbestos (AHERA, 1987; NESHAP, 1990).

1.3.5. **Friable**. "Friable" materials are those materials that, when dry, may be crumbled, pulverized, or reduced to powder by hand pressure, and includes previously non-friable material after such previously non-friable material becomes damaged to the extent that when dry it may be crumbled, pulverized, or reduced to powder by hand pressure (AHERA, 1987).

1.3.6**. Libby Amphibole.** Any mineral belonging to the amphibole assemblage solid-solution series associated with the Libby, MT vermiculite deposit but not limited to only those minerals from that unique location. Includes, but is not limited to, richterite, winchite, tremolite, and actinolite.

1.3.7. **Micaceous Minerals ("Mica").** Hydrous sheet silicates including, but not limited to, vermiculite, biotite, muscovite, phlogopite, and hydrobiotite. Biotite, phlogopite, and hydrobiotite are the source minerals of, and are usually associated with, vermiculite (Bassett, 1959).

1.3.8. **Sprayed-on Fireproofing containing Vermiculite (SOF-V).** A material containing vermiculite, intended to act as a fire-retardant coating when applied to building structures. SOF-V is a surfacing material as specified in 12 NYCRR Part 56 (NYS Industrial Code Rule 56).

1.3.9. Surfacing Material. As defined in 12 NYCRR Part 56 (NYS Industrial Code Rule 56), material that is sprayed-on, troweled-on, or otherwise applies to surfaces (such as acoustical or finish plaster on ceilings and walls, and fireproofing materials on structural members, or other materials on surfaces

for acoustical, fireproofing, or other purposes).

1.3.10 **Vermiculite.** A light brown to gold, hydrous micaceous mineral which expands into accordion-like books upon heating. The expanded product is commonly used in building insulation and soil amendment and may be observed as both single flakes and books (Bassett, 1959).

1.4. **Methodology to Analyze Common Asbestos Samples.** The table below lists many of the sample types commonly submitted to laboratories for bulk analysis. Please note: not all types of samples that may contain asbestos are listed. This table details the appropriate approved method to be used on a specific material type. While the laboratory is ultimately responsible for the accurate determination of which analysis is needed for each sample type (e.g., 198.1, 198.4, 198.6, or 198.8) and for communicating those testing requirements to their client, the testing scheme outlined in Table 1 must be followed.

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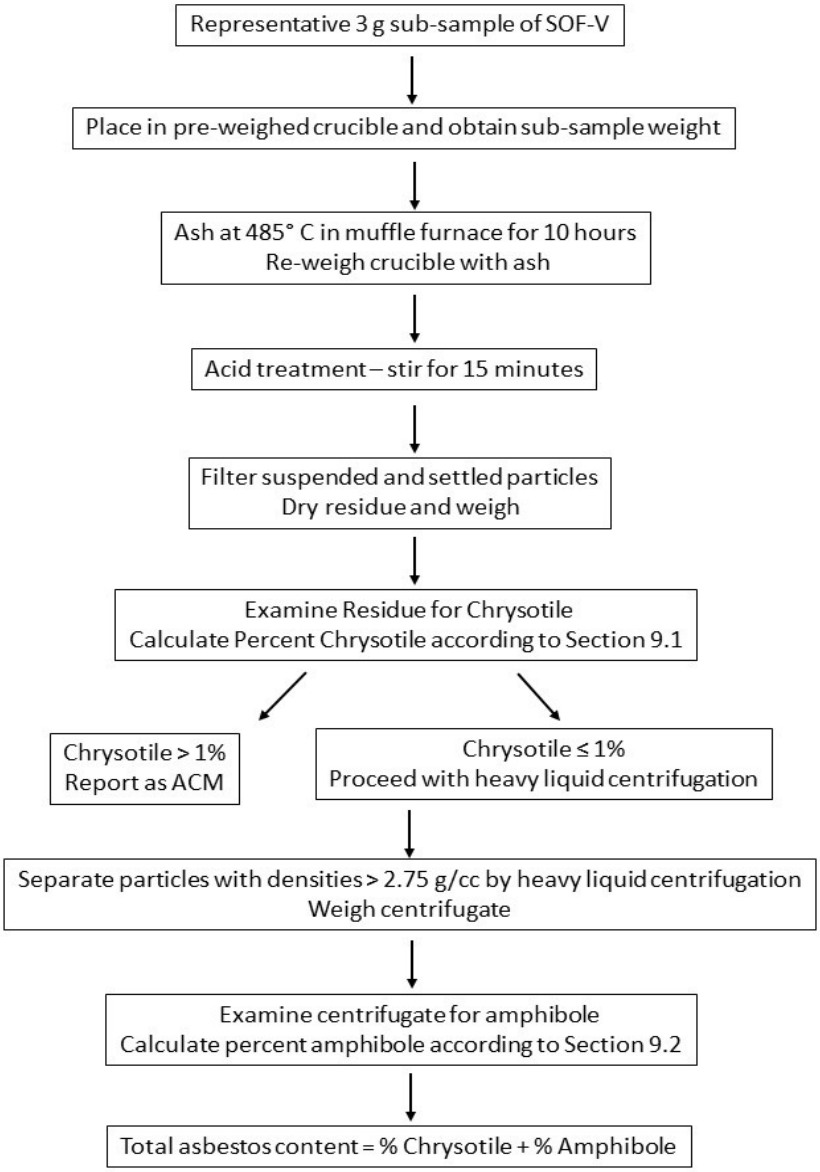
Table 1. Asbestos Sample Types

Material Types that may be analyzed by Item 198.1 (unless NOB material is identified)	**Material Types that must be analyzed by Item 198.6/198.4**	**Material Types that must be analyzed by Item 198.8 or RJ Lee Method 055**
Ceiling Tiles *without* Cellulose	Ceiling Tiles *with* Cellulose	**Surfacing Material containing Vermiculite (SM-V)**
Gypsum Wallboard Joint Compounds	Resilient Floor Tiles	
Wall and Ceiling Plaster	Vinyl Asbestos Tile	
Acoustical Ceiling and Wall Coatings	Mastic	
Sprayed Decorative Coatings (Texture Coats)	Asphalt Shingles	
Asbestos Pipe Packing	Roofing Materials	
Pipe Insulation	Paint Chips	
Duct Wrap	Caulking	
Fiberglass Insulation	Glazing	
Boiler Insulation	Rubberized Asbestos Gaskets	
Furnace Gaskets	Siding Shingles	
House Wrap	**NOB materials (other than SM-V) with < 10% vermiculite**	
Friable materials (other than SM-V) with < 10% vermiculite	**Any material (Friable or NOB other than SM-V) with > 10% vermiculite**	
Surfacing Material (SM) without Vermiculite		

2. **Application**. The method outlined herein is applicable to determine the weight percent concentration of chrysotile and amphibole asbestos in SM-V. These materials types must be prepared by the gravimetric matrix reduction method, in addition to a heavy-liquid separation for amphiboles. The analytical process is depicted in Figure 1 for SOF-V, but can be applied to any other surfacing material.

Figure 1. 198. 8 An example of Analysis of Asbestos in SOF-V.



3. **Equipment and Supplies** The following items shall be available for sample preparation and analysis in laboratories that analyze SM-V:

3.1. HEPA-ventilated, negative-pressure sample preparation work area. This can be a laminar-flow safety cabinet or a similar enclosure that draws all air from the enclosure through a HEPA filter. This should minimize cross contamination and maintain a safe work environment. A flow rate of at least 75 fpm shall be maintained at the opening.

3.2. Low-power (10-45X) stereo binocular microscope with external light source for gross

examination.

3.3. Supplies: glassine paper sheets for examination of samples; scalpel holder and replacement disposable scalpel blades; sampling utensils (dissecting needles and tweezers); cellulose backing pads (optional) may be used to avoid adhesion of the polycarbonate filter to the bottom of the petri dish; Pasteur pipettes; glass rods; aluminum foil; paper towels; small and large spatula; beakers and general laboratory glassware.

3.4. Homogenization equipment:

3.4.1. Mortar and pestle.

3.5. Centrifuge, capable of 3,600 rpm and accommodating at least four 15 mL centrifuge tubes.

3.5.1. Centrifuge tubes. Glass or polypropylene centrifuge tubes, 15 mL capacity to be used in the above centrifuge. The centrifuge tubes must be clear to facilitate removal of heavy liquid without disturbing the pellet of material at the bottom of the tube.

3.6. Plastic Petri dishes (50 mm diameter with lids)

3.7. Muffle furnace capable of sustained operation at 500^{o}C;

3.7.1. Crucibles (bottom and lids) that can withstand 500^{o}C

3.7.2. Instrument or materials capable of calibrating muffle furnace at 485^{o}C:

3.7.2.1. High-temperature thermometer with range to at least 500^{o}C and with readable subdivisions of 5^{o}C or less ***or***

3.7.2.2. Melting-point solids with capability of differentiating 5^{o}C differences between 400^{o}C and 500^{o}C ***or***

3.7.2.3. Independent potentiometer capable of differentiating 5^{o}C differences between 400^{o}C and 500^{o}C.

3.8. Concentrated hydrochloric acid (HCl), reagent grade.

3.9. Heat lamp, slide warmer or drying oven.

3.10. Filtered (0.1 µm) distilled or deionized water

3.11. Textbook or reference book on mineralogy or crystallography, e.g., McCrone 1980, McCrone 1988, Deer, Howie and Zussman, 1966, Shelley 1975.

3.12. Reference materials.

3.12.1. National Institute of Standards and Technology (NIST) SRM 1866a and SRM 1867:

3.12.1.1. Chrysotile.

3.12.1.2. Grunerite (Amosite).

3.12.1.3. Riebeckite (Crocidolite).

3.12.1.4. Glass Fiber.

3.12.1.5. Anthophyllite.

3.12.1.6. Tremolite.

3.12.1.7. Actinolite.

3.12.2. Verified quantitative standards. All laboratories must have at least 4 different SM-V specimens that have been analyzed by an ELAP-certified laboratory (outside lab) using the quantitative method outlined herein. Raw gravimetric data and calculations used to determine percentages of each component (organic, carbonate, insoluble inorganic, and asbestos) must be provided by the outside laboratories and kept on file. Sections of these SM-V must be submitted blindly as routine samples as outlined in Section 13.2.3 to determine the accuracy and precision of the laboratory's current analytical capabilities.

3.12.2.1. Negative (non-ACM) standards. At least two negative standards shall be kept.

3.12.2.2. Positive (ACM) standards. At least two positive standards shall be kept.

3.13. Permanent mount of NIST amosite in n_D=1.680.

3.14. Microscope slides (75mm x 25 mm).

3.15. Cover Glasses (22mm x 22mm).

3.16. Refractive index liquids:

3.16.1. n_D=1.550 high dispersion.

3.16.2. n_D=1.605 high dispersion.

3.16.3. n_D=1.630 high dispersion.

3.16.4. n_D=1.680.

3.16.5. n_D=1.700.

3.16.6. Series of n_D=1.49 through 1.72 in intervals less than or equal to 0.005. This full series is required because of the range of refractive indices exhibited by the different asbestos types in both their natural and altered (heated or acid-stressed) states. High-dispersion liquids may be substituted in the 1.49 through 1.63 range.

3.16.7. Calibration accessory for measuring refractive indices of refractive index liquids. These can be calibrated solids, e.g., glasses, or a refractometer capable of an accuracy of ±0.004.

3.16.8. Calibrated laboratory thermometer with range of 0° to 50° C and readability of ±1° C.

3.17. Marker for labeling slides.

3.18. Polarizing-light microscope equipped with the following:

3.18.1. Sub-stage polarizer.

3.18.2. Analyzer capable of producing a completely black field when privileged direction is oriented perpendicular to that of the sub-stage polarizer. Either the polarizer or analyzer shall be rotatable so that polars can be slightly uncrossed when necessary.

3.18.3. Port @ 45^o to analyzer for wave retardation plate.

3.18.4. 550 nm (first-order red) compensator plate.

3.18.5. 360^o graduated (in 1^o increments) rotating stage.

3.18.6. Illuminator and adjustable diaphragm.

3.18.7. The following objective lenses:

3.18.7.1. Dispersion-staining objective capable of central stop illumination with magnification of approximately 10X (optional).

3.18.7.2. Low-magnification objective (3.2 to 10X).

3.18.7.3. High-magnification, dry objective (30 to 50X).

3.18.8. Eyepiece(s) of at least 8X magnification containing a fixed cross-hair.

3.18.9. Focusable condenser with center-able iris diaphragm capable of completely eclipsing the back-focal-plane image of the central stop.

3.19. Analytical balance with a sensitivity of 0.0001g.

3.20. Magnetic Stirrer.
3.20.1. Teflon coated magnetic stir bars (5 cm length).
3.20.2. Magnet to remove stir bars from flask after stirring completed.

3.21. Sink-Float® Standard. Sink-Float® Standard, density 2.75±0.005 g/cc at 23°C. (Cargille Laboratories, Inc., Cedar Grove, New Jersey 07009) with certification.

3.22. Water Aspirator. Water aspirator, connected to a sidearm vacuum flask, for removal of heavy liquid from centrifuge tubes and collection of the liquid for recovery.

3.23. Erlenmeyer Flasks. Erlenmeyer (conical) flasks, 250 mL, for acid treatment of samples.

3.24. Filtration Assembly

3.24.1. A 47 mm diameter glass vacuum filtration assembly with a glass frit base, Erlenmeyer sidearm vacuum flask, and connecting vacuum hose, for filtration of ashed and acid-treated samples, and centrifugates.

3.24.2. A 25 mm diameter glass vacuum filtration assembly with a glass frit base, Erlenmeyer sidearm vacuum flask, and connecting vacuum hose, for quality assurance testing of recovered heavy liquid.

3.25. Polycarbonate Filters. 0.4 - 0.8 µm pore size, 47 mm diameter (25 mm is allowed when there is a small amount of centrifugate), polycarbonate (PC) filters, for filtration of ashed and acid-treated samples and the centrifugates.

3.26. Porcelain or Glass Buchner Funnel. 240 mL glazed porcelain Buchner or glass funnel for 90 mm diameter filter papers, for recovery of heavy liquid.

3.27. Whatman Filters. Whatman No. 40 cellulose filters, 90 mm diameter, for coarse filtration of used heavy liquid.

3.28. Mixed Esters of Cellulose Filters (MCE).

3.28.1. 0.22 µm porosity, 47 mm diameter mixed esters of cellulose filters, for final filtration of recovered heavy liquid.
3.28.2. 0.22 µm porosity, 25 mm diameter mixed esters of cellulose filters, for quality assurance testing of recovered heavy liquid.

3.29. Desiccator.

3.30. Heavy Liquid. Capable of adjustment to a density of 2.75 g/cc. Either an aqueous solution of lithium metatungstate or sodium polytungstate is suitable.

3.31. Alcohol. Reagent grade Ethanol or Methanol.

3.32. Analysis sheet. Analysis benchsheets and/or other data forms must minimally capture all of the following elements. Example benchsheets are provided in Appendices A and B.

3.32.1. Analyst's signature.

3.32.2. Date of analysis.

3.32.3. Sample identification number.

3.32.4. Gross description of bulk sample (color, homogeneity, texture), tentative identification of all fibers, and confirmation of vermiculite by stereo binocular microscope.

3.32.5. Matrix reduction. This shall include ashing and acid steps used and amount of matrix removed during each step to the nearest mg.

3.32.5.1. Mass of original sub-sample.

3.32.5.2. Mass of post-ashed sub-sample.

3.32.5.3. Mass of post-acid floats.

3.32.5.4 Mass of post-acid sub-sample.

3.32.5.5 Mass of centrifugate.

3.32.6. Entries for each asbestos type identified:

3.32.6.1. Morphology.

3.32.6.2. Refractive index (at λ_o=589.5 nm) parallel to fiber length in specified n_D medium.

3.32.6.3. Refractive index (at λ_o=589.5 nm) perpendicular to fiber length in specified n_D medium.

3.32.6.4. Sign of elongation.

3.32.6.5. Angle of fiber length extinction.

3.32.6.6. Pleochroism and color.

3.32.6.7. Birefringence.

3.32.6.8. Other observations.

3.32.7. Space for recording asbestos points counted per slide preparation and calculation of asbestos percentage.

3.32.8. Final results including:

3.32.8.1. Percentage(s) of chrysotile and amphibole asbestos detected.

3.32.8.2. Total percentage of asbestos.

3.32.8.3. Percentage of organic fraction and water.

3.32.8.4. Percentage of Floats.

3.32.8.5. Percentage of Residue.

3.32.8.6. Percentage of Centrifugate.

3.33. Cotton applicator swabs; may be used to remove material adhered on the upper parts of the centrifuge tube.

4. **Sample Preparation.** Careful sample preparation is a critical step in the analysis. Examine the whole sample using a stereo-binocular microscope at magnifications up to approximately 40x to detect any obvious layers or lack of homogeneity and to confirm that expanded vermiculite is present. Morphologically, vermiculite exhibits accordion-like 'books' after expansion. These 'books' are composed of single platy sheets which may flake off and be observed singly and mis-categorized as "mica". If books are observed, then the single sheets must be considered to be vermiculite, and 198.8 must be used for further analysis.

******If vermiculite is not detected, analysis by Item 198.8 must be terminated. Proceed to analyze the sample utilizing Items 198.1 or 198.6 according to the absence /presence of NOB material, and in accordance to the most current NYS Department of***

Health guidance.***

The EPA and OSHA have clarified how bulk samples that contain multiple layers are to be analyzed and reported (US EPA, 1995; US EPA, 1994a; US EPA, 1994b, OSHA Standards Interpretations 1926.1101). *Layered samples must be handled according to the most stringent of these guidelines.*

4.1. **Preliminary Examination.** Each sample must be examined in its entirety by stereo binocular microscopy. This mandatory stereo binocular microscopic examination will serve three purposes.

4.1.1. Determine the existence of different materials or layers: Confirm the presence of vermiculite. Each discrete material must be prepared, analyzed, and reported separately.

4.1.2. Determine the overall homogeneity of the material: If the material is homogeneous, sub-sampling of sections for preparation and analysis can be at random.

4.1.3. Search for protruding fibers: Fibers protruding from the matrix may be removed for PLM analysis. The identification of asbestos by PLM in this step must be noted on the analysis sheet.

4.2. **Gravimetric Reduction.** The distribution of asbestos in SM-V is often very inhomogeneous. The only way to ensure that the analytical result is representative of the material is to analyze a sufficiently large sub-sample. A minimum weight of 3 grams is required for analysis. The laboratory shall notify clients, (whether internal or external) that a minimum of 10 grams of sample is required to be collected and submitted to the laboratory for analysis and retention. The remaining unused portion of the original sample shall be archived for a period no less than 90 days from the date that the laboratory report is transmitted to the client. Samples shall be submitted dry and as free of extraneous material (paint, rust layers, fragments of cement or encapsulant, etc.) as possible. If insufficient SM-V is available for complete analysis after removal of extraneous material, the sample is not to be tested and the client must be notified regarding why the sample was rejected.

4.2.1. **Removal of Organic Material.** Pre-weigh a crucible with the lid. Lightly crush a representative sub-sample of the fireproofing using an agate mortar and pestle, and place a minimum of 3 grams of the crushed material into the crucible. Weigh the crucible with lid containing the fireproofing sub-sample, to obtain the initial weight of fireproofing to be analyzed.

4.2.1.1. Place the crucible with lid into the muffle furnace, and ash the sample at 485 ± 5°C for a minimum period of 10 hours. Remove the crucible with lid from the muffle furnace, allow it to cool to room temperature in a desiccator, and weigh the crucible with lid.

4.2.2**. Acid Treatment.** Transfer the ashed sub-sample to a 250 mL Erlenmeyer conical flask, add the Teflon coated stirring bar, and also add 100 mL of 2M hydrochloric acid. Add an additional 50 mL of 0.1 µm filtered water. Use a wash bottle with 0.1 µm filtered water to rinse any of the sub-sample attached to the inside surfaces of the flask into the acid solution. Place the flask on the magnetic stirrer, and stir for 15 minutes.

4.2.3. By holding the magnet to the outside of the Erlenmeyer flask, remove the stir bar by sliding it up the side of the flask. Add 0.1 µm filtered water until the surface level is just at the top of the flask. Wait 5 minutes for any floating material to come to the surface. Using a spatula, remove the floating material and transfer it to a pre-weighed plastic petri dish. Using a Pasteur pipette, return to the flask any liquid in the petri dish that was transferred along with the floating material. Add 2 mL of 0.1 µm filtered water to the petri dish to rinse the floats. Remove this water with the pipette and add it to the Erlenmeyer flask. Repeat this rinse at least 3 times, until the water becomes clear and no color remains. Additional rinses may be required. Dry the floating material in the petri dish on a slide warmer or drying oven until stable weight is achieved (less than 3% difference in weight). Record the mass of the petri dish with its contents.

4.3. **Filter the Residue**. Pre-weigh a new petri dish with a 47 mm, 0.4 to 0.8 µm pore size polycarbonate filter. Set up the glass filtration assembly with the pre-weighed polycarbonate filter and connect it to the water aspirator. Transfer the remaining materials (liquids and solids) in the Erlenmeyer flask to the filter reservoir. Completely rinse the flask with 0.1 µm filtered water and add it to the filter reservoir. After the filtration has completed, rinse the filtered material with at least 50 mL of 0.1 µm filtered water, and allow the filtration to complete. Repeat the rinse at least 2 times to ensure that all soluble materials have been removed from the residue.

4.3.1. Remove the filter and residue from the glass frit base. The deposit on the filter will still be a substantial proportion of the original sub-sample weight (typically 20% to 35%), and will be several millimeters in thickness. With the vacuum still applied, detach the residue from the inside edge of the reservoir using a small spatula. When complete, the filter reservoir can be lifted, leaving the filter and the deposit on the filtration base. After the vacuum is turned off, the filter and residue can then be transferred to the pre-weighed petri dish. If any residue still adheres to the inside of the filter reservoir or base, remove as much as possible with a spatula and transfer it to the petri dish. Dry the filter and residue on a slide warmer or in a drying oven until stable weight is achieved (less than 3% difference in weight). Record the mass of the petri dish with its contents.

5. **Sample Analysis**

5.1. **Identification.** It is expected that analysts using this method are competent in the

identification of asbestos by PLM and can refer to texts such as McCrone (1980, 1988) for assistance in identification. All fibrous components in each residue shall be positively identified. Typical properties of regulated asbestos are outlined in Table 2 at the end of Item 198.8. Deviations from these properties are sometimes seen for asbestos from atypical ores or, more frequently, for asbestos that has been altered chemically or thermally (Laughlin and McCrone, 1989). Materials that commonly interfere with the identification of asbestos are detailed in Section 2 of the US EPA's Test Method (Perkins and Harvey 1993). For asbestos fibers identified in each sample, the following criteria are required on the analysis sheet:

5.1.1. Morphology.

5.1.2. Refractive index (measured at λ_o=589.5 nm) fiber length *and* fiber width. This shall be a numerical value (±0.004) that can be determined by the Becke line method or by use of dispersion staining tables (e.g., McCrone 1989, Su 1994). Laboratory temperature must be measured using the calibrated laboratory thermometer and used in calculating refractive index.

5.1.3. Sign of elongation.

5.1.4. Pleochroism and color.

5.1.5. Extinction angle along fiber length.

5.1.6. Birefringence.

5.1.7. Additional observations are required for difficult samples.

5.2. **Quantitation**. Accurate quantitation is most critical at the 1 percent level, the level differentiating ACM vs non-ACM. Because the US EPA's initial ACM definition was based on weight and because of PLM's limitation to determining areal percentage, the National Emission Standards for Hazardous Air Pollutants (NESHAP, 1990) rule defines "friable asbestos material" and "nonfriable [sic] ACMs" as "containing more than 1 percent asbestos as determined using" the EPA (1982) interim PLM method. While the EPA PLM method analyzes on an areal basis, it also allows removal of matrix materials and "requires a correction for percent weight loss". Thus weight percentage and area percentage determinations can be combined during analysis. The NESHAP preamble (55 *FR* 48410) includes an important discussion of quantitation of ACMs.

The analyst is encouraged to review references in Section 14 for useful strategies in the identification of asbestos. High-contrast Becke lines can enhance detection of fine fibers. Lower or higher RI liquids (solvents or Cargille oils) may be used to increase Becke-line contrast and aid in detection and quantitation of fibers. Asbestos type(s) shall be positively identified in appropriate media according to all criteria in Section 5.1.

Matrix losses shall be gravimetrically tracked at each preparation step (Section 4.1 through 4.3 and Section 7). The determination of asbestos percentage versus the percentage of remaining insoluble inorganic matrix material shall be accomplished using 8 coverslip preparations. This shall be done by a 400 point-counting method.

5.2.1. **Point Counting Criteria**. A point is a discrete point or the intersection of two mutually perpendicular lines in the eyepiece reticle. A nonempty point is the visual superposition of a point over any material in the slide preparation. A nonempty point shall be categorized as a specific asbestos variety, as a specific non-asbestos fiber type or as non-fibrous material (see Section 5.1 for identification criteria), while empty points are those points that lie over areas containing no materials. Ideally, slide preparations should contain approximately 50% nonempty points. Moving to new fields of view shall be done at random, with the analyst looking away temporarily while moving the slide. The slide shall never be deliberately moved to preferred fields of view under the reticle. If the point lies over an area where particles are heavily clumped, the analyst should move the slide to a new field to avoid attempting to count multiple layers under a point. For the occasional superposition of a point over two particles, the analyst should count both particles as separate nonempty points.

5.2.2. **Trace Levels of Asbestos**. If asbestos appears in a field of view but does not lie directly under a point, the analyst shall note this on the analysis sheet. If the analyst suspects that, after a thorough examination, asbestos is present but none is detected during the point-count analysis, the analyst shall retrieve the residue or centrifugate, remove any suspicious fibers, mount them in an appropriate medium, and determine their identity. If the fibers are confirmed as asbestos, this should be noted on the analysis sheet. Although these observations will not be used for quantitation, they will be incorporated into the final report to warn about potential false negatives.

6. **PLM Examination for Chrysotile**.

Point counting shall be done on the PLM, usually with the slide between crossed polars and with a first-order red compensator inserted in the 45^{o} port above the slide. In some situations where extremely fine asbestos fibers are present, it may be preferable to analyze the sample between *slightly* uncrossed polars without the compensator. Other situations may warrant point counting in a dispersion-staining mode. All point counting shall be done at 100x magnification although it will be advantageous at times to switch to higher magnification(s) for enhanced visualization of identification criteria. Appendix C shows PLM micrographs that illustrate the appearance of chrysotile fibers and bundles in a residue from a typical SOF-V that was spiked with 0.6% chrysotile.

6.1 **Slide Preparation.** Prepare eight 75 mm x 25 mm microscope slides from the residue using a high dispersion liquid of refractive index 1.630 or 1.680, and apply a whole 22 mm x 22 mm cover glass to each.

6.2 **Identification of Chrysotile fibers.**

6.2.1. Using crossed polars with a 550 nm compensator plate inserted, thoroughly scan the entire area with overlapping fields of view on each of the 8 slides to determine if structures morphologically consistent with chrysotile are present. In some situations where extremely fine asbestos fibers are present, it may be preferable to analyze the sample between *slightly* uncrossed polars without the compensator.

6.2.2. If no such structures are detected, assign zero chrysotile points and 50 occupied points to each of the 8 slides.

6.2.3 If any structures morphologically consistent with chrysotile are detected, prepare at least 1 additional microscope slide using a 1.550 refractive index oil, and positively identify a minimum of four structures according to Section 5.

6.3 **Quantitation of Chrysotile.**

6.3.1 Use the original 8 slides (using the 1.630 or 1.680 oil) to perform a 400 point count with 50 non-empty points per slide in accordance with section 5.2.

6.3.2 Point counting shall be performed using a fixed cross-hair. A Chalkley reticule shall not be permitted with this method.

7. **Determination of Amphibole Asbestos Concentration Using Heavy Liquid Centrifugation.**

7.1. Weigh the remaining residue in the petri dish. ***NOTE: The residue is hygroscopic.*** If the residue has been exposed to room air for more than 1 hour, the residue must be placed back into the drying oven or desiccator for a minimum of 1 hour before weighing.

7.2. Place petri dish on top of a piece of aluminum foil (e.g., 15 cm x 15 cm square) to capture any reside that may be disturbed during the transfer.

7.3. Use a scalpel to cut the residue into pieces that will fit into the centrifuge tubes. Transfer half of the residue into each of the two centrifuge tubes.

7.4. Transfer any material remaining on the petri dish, lid and aluminum foil into the two centrifuge tubes.

7.5. Calibrate heavy liquid as described in Appendix D prior to use.

7.6. **Disperse the Residue in Heavy Liquid.** Add 10 mL of density-adjusted heavy

liquid to each of the centrifuge tubes. Using a glass rod, disperse the residue in the liquid by macerating the solids between the inside of the centrifuge tube and the rod. Add additional heavy liquid to bring the liquid level up to 2 cm from the top of each centrifuge tube. Disperse the solids throughout the liquid using the glass rod.

7.7. **Centrifuge the Tubes and Wash the Heavy Fraction**. The required time for centrifugation depends on the dimensions and rotation speed of the particular centrifuge in use. Refer to Appendix E for the required centrifuge times for amphiboles in heavy liquid with different dimensions and rotation speeds. The majority of the weight of amphibole in vermiculite is present as relatively large fragments. Use a centrifuge time that is sufficient for sedimentation of all particles larger than 5 µm diameter and with a minimum density of 3.0 g/cc.

7.7.1. Place the centrifuge tubes in opposite positions in the centrifuge, and operate the centrifuge at maximum speed for the calculated time. (For example, the dimensions of the centrifuge and rotation speed in the example shown in Appendix E, the required time is 5 minutes.)

7.7.2. After centrifugation, remove each of the tubes from the centrifuge. There will be a small pellet of solids at the bottom of each tube, and a much larger amount of floating material will accumulate as a solid plug at the top of the heavy liquid. Using the glass rod, macerate the floating fraction against the inside of each tube, and re-disperse the material in the heavy liquid, without disturbing the pellet of solids at the bottom of the tube. Centrifuge the tubes for another 5 minutes.

7.7.3. Repeat the dispersal and centrifugation one more time as described in 7.7.2.

7.7.4. Using a small spatula, remove and discard as much of the solid plug of floating material as possible from the top of each centrifuge tube.

7.7.5. Aspirate as much of the heavy liquid as possible from each tube, taking care to not disturb the pellet of particulate material at the bottom. Save the collected liquid for recovery and reuse (refer to Appendix G).

7.7.6. There will be some residual floating material adhering to the upper parts of the centrifuge tubes. Remove this adhering material from the upper parts of the centrifuge tubes using a cotton applicator swab. It may be necessary to wet the cotton swab with 0.1 µm filtered water to facilitate the removal of the adhering material.

7.8. **Wash centrifugate to remove heavy liquid.** Direct a jet of 0.1 µm water from a wash bottle at the pellet of particulate in each centrifuge tube. This action should disperse the pellet in the water. Redistribute the pellet in 5 mL of 0.1 µm water.

7.8.1. Centrifuge the tubes for 1 minute.

7.8.2. Remove tubes and pour out the liquid, taking care to not to disturb the pellets at the bottom of the tubes. Repeat the washing and centrifugation steps 2 more times to remove traces of the heavy liquid.

7.8.3. Weigh a petri dish containing a 47 mm, 0.4 to 0.8 µm pore size, polycarbonate filter (a 25 mm filter may be used when there is a small amount of centrifugate).

7.8.4. Set up the filtration assembly with the polycarbonate filter and connect it to the vacuum.

7.8.5. After the final centrifugation, transfer the contents of the both centrifuge tubes to the filter reservoir.

7.8.6. Rinse the centrifuge tubes with 0.1 µm water and add it to the filter reservoir.

7.8.7. Apply vacuum to the filter assembly. After the filtration is completed rinse the filtered material with at least 50 ml of 0.1 µm water and allow filtration to complete.

7.8.8. Remove the polycarbonate filter with the centrifugate and place it in to the petri dish on a slide warmer or drying oven until stable weight is achieved (less than 3% difference in weight). Record the mass of the petri dish with its contents.

8. **PLM Examination for Amphibole Asbestos**. For vermiculite that originated from Libby, the centrifugate will contain variable amounts of fiber bundles that exhibit magenta/gold (parallel) and blue (perpendicular) dispersion staining colors; often with a non-zero extinction angle. Reliable and routine discrimination between the various amphiboles present in vermiculite from Libby is not possible by PLM; therefore the fibers shall be identified as "amphibole asbestos". The optical properties for 4 fibers/fiber bundles must be recorded as described in Section 5. Examples of the appearance of amphibole asbestos fiber bundles are shown in Appendix F. It is important to note that fiber bundles of amphibole asbestos, when present as contamination in vermiculite, often have attached fine particulate that limits the ability of the analyst to observe dispersion staining colors. The fiber bundle shown in Appendix F (C) is an example of such a bundle. Appendix F (E) is an example in which identification of the fiber bundle by dispersion staining is compromised by the overlapping plate of vermiculite.

8.1. **Slide preparation.**
Prepare eight 75 mm x 25 mm microscope slides from the centrifugate using a high

dispersion liquid of refractive index 1.630, and apply a whole 22 mm x 22 mm cover glass to each.

8.2. **Amphibole Identification.**

8.2.1. Scan the whole area of each of the 8 slides at a magnification at 100X in accordance with Section 5. Many of the fragments of amphibole asbestos that may be present are similar in size to the flakes of vermiculite. Most of the mass of any amphibole asbestos will be represented by these large fragments.

8.2.2 If no amphibole asbestos fibers or fiber bundles are observed during these examinations, assign 50 occupied points and zero asbestos points to each of the 8 slides. If amphibole asbestos is detected, discontinue the scan and proceed to point counting.

8.3. **Amphibole Quantitation.**

8.3.1 If any amphibole asbestos is detected, perform a 400 point count. Point counting shall be carried out using a single point cross-hair. A Chalkley reticule shall not be permitted with this method.

8.3.2. Calculate the concentration of amphibole asbestos in the centrifugate and the concentration of amphibole asbestos in the original sample. An example of the required calculation procedure is shown in Appendix B (where the shaded cells represent the required input data for the calculations) and described in Section 9.

9. Calculations. Calculations are performed for chrysotile and amphibole asbestos separately. The following calculations use the same values as those in Appendix B.

9.1 **Calculation for Chrysotile.** For example, 8 slides were prepared from the residue after ashing and acid treatment. During point counting on all 8 slides, a total of 13 chrysotile points were counted and 387 non-chrysotile points (including any amphibole structures observed) were counted.

The percentage of chrysotile in the original sample is calculated by:

% Chrysotile = (PAM / OM) x (CP / CTP) x 100%

Where:
PAM = Mass of residue after furnace and acid treatments (g) (eg. 0.7645 g)
OM = Mass of original sub-sample (g) (eg. 3.0865g)
CP = Number of chrysotile points in point count of residue (eg. 13)
CTP= Total number of nonempty points in chrysotile point count (eg. 400)

Thus:
% Chrysotile = (0.7645g/ 3.0865g) x (13/400) x 100%
= 0.8050%

9.2. **Calculation for Amphibole asbestos.** For example, 8 slides were prepared from the centrifugate remaining after ashing, acid treatment and heavy liquid centrifugation. During point counting on all 8 slides, a total of 58 amphibole asbestos points were counted and 342 non-amphibole points (including any chrysotile structures observed) were counted.

The percentage of amphibole asbestos in the original sample is calculated by:
% Amphibole Asbestos = (PHM / CR) x (PAM / OM) x (AP / ATP) x 100%

Where:
PHM = Mass of centrifugate after heavy liquid centrifugation (g) (eg. 0.1317g)
PAM = Mass of residue after furnace and acid treatments (g) (eg. 0.7645g)
OM = Mass of original sub-sample (g) (eg. 3.0865g)
CR = Mass of residue post-chrysotile analysis (g) (eg. 0.7279g)
AP = Number of amphibole asbestos points in point count of centrifugate (eg. 58)
ATP = Total number of nonempty points in amphibole asbestos point count (eg. 400)

Thus:
% Amphibole asbestos = (0.1317g / 0.7279g) x (0.7645g / 3.0865g) x (58 / 400) x 100%
= 0.6498%

9.3. **Calculation of Total Asbestos Content.**
Total percent asbestos = C + A
where:
C = Percentage of chrysotile calculated in 9.1
A = Percentage of amphibole asbestos calculated in 9.2

Thus:
Total Asbestos in original sample = 0. 0.8050% + 0.6498%
= 1.4548%

9.4. **Rounding Rules.** The percentage of asbestos must be rounded off to two significant digits. Results ending in 5 or greater are rounded up. Results ending in 0 to 4 are rounded down. For example, a total asbestos result of 1.05% is rounded to 1.1%. A total asbestos result of 1.01% is rounded to 1.0%.

10. **Analytical Records**. Detailed records shall be kept of all phases of analysis. Analysis

sheets that include all the data required in Section 3.32 shall be filled out completely, signed and dated by analyst.

11. **Test Reports**. Reports to clients shall include at least the following:

11.1. **Client**. Identify name and address.

11.2. **Sample Identity**. The identification number (Section 13.1.5.1.3.) assigned by the laboratory shall be clearly cross-referenced to information provided by the client or collector (field identification number, location - Section 13.1.5.1.2.) for each sample.

11.3. **Date of analysis**.

11.4. **Identity of analyst**.

11.5. **Analytical Results**. The following information shall be reported for each sample:

11.5.1. Color.

11.5.2. Presence (or absence) of chrysotile and/or amphibole asbestos, and total percentage of asbestos shall be reported as follows:

11.5.2.1. "No asbestos detected" - for samples that contained no asbestos points and no trace asbestos (Section 5.2.2) as confirmed by PLM.

11.5.2.2. "Trace **(fill in type(s))** asbestos detected." - for samples that contained 0 asbestos points out of 400 (or more) nonempty points but did contain asbestos positively identified by PLM (Section 5.2.2).

11.5.2.3. " **(fill in type)** asbestos detected at ____%" - for each type of asbestos for which asbestos point(s) were counted. Percentage must be rounded off to two significant digits.

11.5.2.4. " ____% Total Asbestos" - sum from all types reported in (Section 9.3.) Percentage must be rounded off to two significant digits.

11.5.3. Presence of other fibrous materials observed.

11.5.4. Copies of the laboratory's benchsheets shall be included as part of the final test report package.

11.6 **Homogeneity**. For samples with obvious layers, the summary shall include results as specified in Section 11.5 for each layer. Compositing of layers into a single result is no longer allowed, except for joint compounds in certain cases (U.S.E.P.A. 1994b).

12. **Precision and Accuracy**.

12.1. **Precision.** Theoretically, the point-count method should yield a relative standard deviation no better than 50% at a composition of 1% asbestos when performed with 400 points. However, depending on matrices within a bulk sample, the point-count method (when performed with 300 or more points) has yielded relative standard deviations of 25% or less at compositions between 1 and 3 percent asbestos (Perkins 1989). Similar results were derived from synthetic bulk samples with formulated weight compositions; relative standard deviations ranged from 24 to 45% for replicate samples containing 5 to 7% chrysotile and amosite (Webber et al. 1990). Certain matrices (e.g., chrysotile in vermiculite or cellulose) tend to increase analytical variability.

12.2. **Accuracy**. The point-count method is usually more accurate than the visual estimation method at quantitating asbestos concentrations in bulk samples, especially at low concentrations (Perkins 1989, Perkins 1990, Webber et al. 1990, Harvey et al. 1991). Nonetheless, there are material combinations that seem to cause biases (e.g., the presence of cellulose causes underestimation of chrysotile while amosite in plaster is usually overestimated). In even these instances, however, point-count results are usually closer to actual weight percentages than visual-estimation results.

13. **Quality Assurance**

13.1. **Quality-Assurance Manual**. The laboratory's QA manual can be devoted to asbestos analysis or it can be a larger manual comprising many types of analyses. In either case, the QA manual shall include at least the following and shall be in conformance with the general ELAP requirements for quality manuals:

13.1.1. **Quality Assurance Responsibility**. A single individual shall be designated as responsible for overseeing quality assurance. This includes updating and controlling distribution of the laboratory's quality-assurance manual, performing at least monthly reviews of analytical quality control and contamination control and resolving any deficiencies.

13.1.2. **Analytical Method.** The laboratory's implementation of the SM-V method shall be explicitly detailed in this manual. If a copy of an externally published method (e.g., this document) is used, then it shall be customized to include only those options in the approved method that are actually utilized by the laboratory.

13.1.3. **Analytical Quality Control.** The manual shall describe a systematic method of submitting quality-control samples including intra-analyst, inter-analyst, standards,

proficiency-testing and inter-laboratory samples so that analysts are unaware of the sample's true identity.

13.1.4. **Sample Control.** The manual shall describe all aspects of SM-V sample handling from sample receipt to sample disposal. Criteria for acceptance and rejection of received samples (e.g. broken containers, too-small samples) and for safe handling shall be defined. Samples shall be retained in secure areas (similar to areas used to store evidentiary material) for at least 90 days after a final report of results is sent to the client. Samples may be returned to the client at the client's request at any time. Procedures for safe disposal of asbestos (in compliance with federal and local regulations) shall be detailed and records of such disposals shall be kept.

13.1.5. **Recordkeeping.** The laboratory shall maintain a recordkeeping system as specified in its QA manual. This shall define provisions to ensure the secure storage of records for at least five years. Records, whether they be hardcopy or computer files, shall be easily accessible and shall include:

13.1.5.1. **Sample Accessioning**. Each sample shall pass through an accessioning process that documents:

13.1.5.1.1. **Client**. This should include name, address, phone number and name of contact person.

13.1.5.1.2. **Client Sample Identification**. This should include the identification characteristics provided by the client, e.g., identification number, collection site, etc.

13.1.5.1.3. **Laboratory Sample Identification**. A unique laboratory sample identification number shall be assigned to each sample.

13.1.5.1.4. **Date and Time of Receipt.**

13.1.5.1.5. **Chain of Custody.**

13.1.5.1.6. **Condition of Sample. (Accept/Reject)**

13.1.5.1.7. **Type of Sample**. This should place the sample in one of several bulk sample categories, e.g., friable, floor tile, sprayed on fireproofing, etc.

13.1.5.2. **Analytical Quality Control**. All results of analytical quality control activities shall be recorded in an orderly fashion.

13.1.5.3. **Equipment and Supply Records**. Records shall be kept for maintenance, calibration, replacement and repair of pertinent equipment and supplies. For major pieces of equipment (microscopes, hoods, muffle furnaces,

analytical balances) these records shall include manufacturer, model and serial numbers, major components, calibration and maintenance/service information and location of manuals.

13.1.5.4. **Contamination Control**. See Section 13.3.

13.1.5.5. **Calibration**. See Section 13.4.

13.1.5.6. **Personnel**. See Section 13.6.

13.1.5.7. **Test Reports**. See Section 11.

13.1.6. **Staff Training Programs**. The Laboratory Director is responsible for continued in-house training of analysts. Each analyst shall receive formal training in proper identification and quantitation of asbestos in bulk samples. This can be achieved by sending the analyst to a 5-day course at a recognized PLM institute or by an in-house training course with a detailed and extensive curriculum equivalent to that at recognized institutes. The course shall include formal training in the theory of mineral analysis by PLM and hands-on analysis of all asbestos types and common fiber types. This formal training shall be followed by an in-house apprenticeship during which performance is carefully monitored and documented to show increasing competence to the point where the analyst can work independently within the laboratory's QA framework.

13.2. **Analytical Quality Control**. At least 10% of a laboratory's SM-V analyses shall be re-analyzed as part of the laboratory's QC program. Selection of samples for quality-control (intra-analyst, inter-analyst, inter-laboratory, or reference) analyses shall be semi-random so that the analyst performing the original analysis is not aware that the sample will be reanalyzed. Furthermore, the second analyst shall not know the results of the original analysis. These QC data shall be routinely assessed to evaluate the precision and accuracy of each analyst and to identify and correct areas of analytical weaknesses. These QC samples shall be routinely resubmitted for analytical quality control according to the method detailed in Section 13.1.3. QC reanalysis shall include complete re-preparation (including gravimetric reduction) of slides from the original sample. All QC results shall be documented in a QC notebook or on appropriate analysis sheets. Procedures for resolving analytical discrepancies shall be defined and details of resolved discrepancies shall be recorded. Discrepancies include classification differences (ACM vs. non-ACM), identification differences (e.g., chrysotile vs. amosite) and substantial quantitation differences, as specified below. Monthly summaries shall be compiled for each analyst.

One of QC's primary functions is the timely detection and correction of deficiencies in an analytical system. QC is not an optional activity to be carried out at the convenience of the laboratory or to be postponed when sample loads are heavy. ELAP-certified laboratories **shall** perform PLM QC concurrent with sample load and

shall evaluate these QC results before sending written reports to clients.

13.2.1. **Intra-Analyst QC**. At least 1 out of 50 samples shall be reanalyzed by the same analyst. Relative difference (R) values shall be calculated for each pair of re-analyses and shall be compiled and statistically evaluated for that analyst, comparing his/her QC result to his/her original result for that same sample. The intra-analyst R values are absolute values and are calculated using

$R = | (A-B) / ((A+B)/2) |$

where

A = First result from the analyst being checked

B = Second result from same analyst for same sample

Intra-analyst results will require additional reanalysis, possibly including another analyst, to resolve discrepancies when classification (ACM vs. non-ACM) errors occur, when asbestos identification errors occur, or when R is greater than 1.0.

Record: Sample, date(s) of analyses, analysts' signatures, both results, R value, reason(s) for and resolution(s) of disagreement(s). R control charts shall be updated monthly for each analyst monitoring intra-analyst precision. These charts shall include all R values from at least the three previous months.

13.2.2. **Inter-Analyst QC**. At least 1 out of 15 samples shall be reanalyzed by another analyst. R values shall be calculated for each pair of re-analyses and shall be compiled and statistically evaluated for each analyst, comparing his/her result to a QC result for that same sample from another analyst. R value are calculated using:

$R = (A-B) / ((A+B)/2)$

where

A = Result from the analyst being checked

B = Result from other analyst for same sample

Inter-analyst results will require additional reanalysis, possibly including another analyst, to resolve discrepancies when classification (ACM vs. non-ACM) errors occur, when asbestos identification errors occur, or when R is greater than 1.0 or less than -1.0.

Obviously single-analyst laboratories will not be able to meet this requirement. Instead, they shall perform **intra-analyst** analyses on 1 out of **every 10 samples**.

Record: Sample, date(s) of analyses, analysts' signatures, both results, R value, reason(s) for and resolution(s) of disagreement(s). R-bar control charts shall be updated monthly for each analyst monitoring both intra-and inter-analyst precision. These charts shall include all R values from at least the three previous months.

13.2.3. **Standard/Reference QC**. At least 1 out of 100 samples shall be a verified quantitative standard (Section 3.12.2) that has been routinely resubmitted to determine analyst's precision and accuracy. Results should be displayed on x-bar charts to keep track of each analyst's accuracy and precision.

13.2.4. **Inter-laboratory QC**. The laboratory must participate in round-robin testing with at least one other ELAP-certified lab. For laboratories with more than one bulk-sample analyst, samples must be sent to this other lab at least four times per year or at the rate of 1 sample in 500 routine samples (whichever is less). For single-analyst laboratories, at least 1 sample in 500 routine samples must be sent to this lab. These samples must be samples previously analyzed as QC samples. Results of these analyses must be assessed in accordance with QC-outlier criteria detailed in the lab's QA manual. At the very least, the QA manual must address misclassifications (false positives, false negatives) and misidentification of asbestos types.

13.3. **Contamination Control**.

13.3.1. **Prevention**. The laboratory shall detail its methods for preventing cross contamination of equipment, supplies and reagents. Much of this will be careful cleaning of work area, equipment and supplies. Intensity and frequency of this effort should be based on experience gained through any contamination detected as described in 13.3.2.

13.3.2. **Monitoring**. The laboratory shall have a documented routine procedure for monitoring contamination of laboratory equipment, supplies and work stations and for resolving contamination problems when discovered. If any asbestos is detected, the source of contamination shall be traced and the problem resolved to prevent recurrence. Any of the previous samples that may have had results affected by the contamination shall be reanalyzed and the client notified of any revisions to reported values. Detailed records of monitoring shall be maintained. At least one non-ACM SOF-V shall be prepared and analyzed with every 20 samples analyzed. This non-ACM shall go through the full preparation and analysis regimen for the type of analysis being performed.

13.4. **Calibration**. All calibrations listed below (unless otherwise noted) shall be performed under the same analytical conditions used for routine asbestos analysis. Frequencies stated below may be reduced to "before next use" if no samples are analyzed after the last calibration period has expired. Likewise, frequencies shall be increased following non-routine maintenance or unacceptable calibration performance. The information must be recorded in a bound notebook, three-ring binder, spread sheet, or an equivalent, permanent record. Refer to Certification Manual Item 231 for additional information on QA requirements and records for equipment. At a minimum, the following items and associated records are to be included.

13.4.1. **Refractive Index Media**. The refractive index medium (oil or solid) used to prepare slides shall be calibrated to within 0.004 using certified refractive-index solids or a refractometer. This shall be performed when the original container is first opened for use and thereafter at three-month intervals.

Record: Date, nominal refractive index, measured refractive index, temperature, and analyst's initials

13.4.2. **Laboratory Thermometer**. The laboratory thermometer must be calibrated to a NIST-traceable standard annually to ±1° C within a temperature range of 20° to 30°C.

Record: Date, thermometer ID, calibration temperature, correction factor, and analyst's initials

13.4.3. **PLM Alignment**. The PLM shall be aligned daily, or next use, to achieve illumination as close to Köhler illumination as possible and centered through the sub-stage condenser and iris diaphragm. The stage's rotation axis shall be centered with the appropriate objectives. Analyzer and polarizer shall be rotated to maximum extinction with each other and their privileged directions shall be oriented parallel to the lines in the fixed ocular cross hairs (or grid) and aligned at 45^{o} to the accessory port.

Record: Date, check-off for rotation centering, axial illumination, full extinction and crosshair alignment fixed in the polarizer's privileged direction, and analyst's initials

13.4.4. **Refractive-Index Colors**. Dispersion-staining or Becke-line colors shall be determined monthly from the permanent 1.680 mount of amosite (Section 3.13). The source of any deviations shall be located and corrected.

Record: Date, colors or wavelengths perpendicular and parallel to length, and analyst's initials

13.4.5. **Analytical Balance.**

13.4.5.1. Analytical balances should be serviced by a qualified service organization annually.

Record: List of balances including date of service and Certificate of Weight Traceability, service organization sticker with date of service fixed to each balance, and calibration data

13.4.5.2. Analytical balances shall be checked in the working range daily with NIST traceable class S weights. The range selected should reflect the routine use of the balance and the actual class S weights used should test the optical scale at mid-point.

Record: Date, target weight, measured weight, and analyst's initials

13.4.6. **Muffle Furnace.** Temperatures on external meters (either direct-temperature

displays or graduated potentiometers) shall be calibrated quarterly (Section 3.7.2). This shall be a three-point calibration covering a temperature range of at least 450° to 500°C. If a thermometer is used for calibration, the thermometer bulb should be immersed in a sand bath.

Record: Date, target temperature, measured temperature, and analyst's initials

13.4.7. **HEPA-Ventilated Sample Preparation Enclosure.** Flow rate at enclosure opening shall be measured twice annually to the nearest 5 fpm. Flow rate shall not be less than 75 fpm.

Record: Date, flow rate, and analyst's initials

13.4.8. Heavy Liquid. Calibration of the heavy liquid shall be completed before initial use and after each recovery as per Section 7.5 and Appendix D using a certified standard.

Record: Date, initial or recovered state, calibrated density, Lot number, analyst's initials.

13.6. **Personnel.** The laboratory shall assure that all analysts are competent to perform PLM analysis of asbestos in bulk samples. Analysts shall be familiar with the theory of dispersion staining and the measurement of refractive indices by the Becke line technique and be able to apply these. A personnel file shall be maintained for every analyst and shall include:

13.6.1. **Resumé.** Each resumé shall include formal education, experience and other pertinent information.

13.6.2. **Training.** Both classroom and in-house training shall be detailed to demonstrate the analyst's competence in performing independent analysis.

13.6.3. **Job Title.** A job title shall be defined that specifies responsibilities and laboratory assignments.

13.6.4. **QC Records.** Details and summaries of results of QC analyses shall be updated at least monthly. Accuracy shall be determined from the standard/reference samples while precision shall be determined from intra- and inter-analyst R values.

13.6.5. **Deficiency Resolutions.** Details of noted deficiencies and steps taken to resolve these shall be included in the personnel folder.

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Appendix A. SM-V Sample Analysis Sheet

Analyst:_____ **Analysis Date:**_____ **Sample ID:**_____

STEREOBINOCULAR MICROSCOPY:
Color:_____ **Texture:**_____ **Homogeneity:**_____ **Homogenization:**_____
Probable Fibers:_____ **Remarks:**_____

POLARIZED-LIGHT MICROSCOPY: • Condenser Focused • Condenser Centered • Rotation Centered • Polars Extinct • Crosshair Privileged

IDENTIFICATION:

Morphology	Refractive Index		Sign of Elongation	Extinction Angle	Pleochroism /Color	Birefrin-gence	Other	Identity
	⊥	∥						

Remarks:_____

QUANTITATION: 400 Point Counts

Chrysotile

Slide 1		Slide 2		Slide 3		Slide 4		Slide 5		Slide 6		Slide 7		Slide 8	
Chry	Other	Chry	Other	Chry	Other	Chry	Other	Chry	Other	Chry	Other	Chry	Other	Chry	Other

Amphibole Asbestos

Slide 1		Slide 2		Slide 3		Slide 4		Slide 5		Slide 6		Slide 7		Slide 8	
Amph	Other	Amph	Other	Amph	Other	Amph	Other	Amph	Other	Amph	Other	Amph	Other	Amph	Other

CALCULATIONS:

COMPONENT	Chrysotile	Amphibole	Other	
Points Counted (PC)				
Total Points Counted (TP)				
Percentage (100*PC/TP)				

Verified:_____ **Date:**_____ **Remarks:**_____

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Appendix B. Determination of Asbestos in Surfacing Material containing Vermiculite

SAMPLE:	221B Baker Street, 10th Floor	**SAMPLE NO:**	MHS12345-05
	New York	**DATE:**	April 11, 2014
	Sample 05	**ANALYST:**	MLK

INITIAL WEIGHTS		**Comments**
Weight of Crucible	18.6948	
Weight of Crucible + Sub-Sample	21.7813	
Weight of Sub-Sample	3.0865	
ASHING		
Weight of Crucible + Ash	21.5372	
Weight of Ash	2.8424	
Weight Loss During Ashing	0.2441	
Weight Percent Organic and Water	7.9086	
ACID TREATMENT/FLOTATION		
Weight of Dish for Floats	3.8699	
Weight of Dish + Floats	4.1873	
Weight of Floats	0.3174	
Weight Percent Floats	10.2835	
Weight of Dish + Filter for Residue	3.8162	
Weight of Dish + Filter + Residue	4.5807	
Weight of Residue	0.7645	
Weight Loss During Acid Treatment	1.7605	
Weight Percent Acid-Soluble Materials	57.0387	
Weight Percent Residue	24.7692	

PLM EXAMINATION OF RESIDUE (CHRYSOTILE)			**Chrysotile Point Counts (Chrysotile/Other)**					
Point Count:	Number of Occupied Points	400	Slide 1:	1	49	Slide 5:	1	49
	Number of Chrysotile Points	13	Slide 2:	1	49	Slide 6:	2	48
			Slide 3:	1	49	Slide 7:	2	48
PERCENT CHRYSOTILE IN SAMPLE		**0.8050**	Slide 4:	3	47	Slide 8:	2	48

HEAVY LIQUID CENTRIFUGATION	
Weight of Dish + Filter + Balance of Residue	4.5441
Weight of Balance of Residue	0.7279
Weight of Dish + Filter for Centrifugate	3.7047
Weight of Dish + Filter + Centrifugate	3.8364
Weight of Centrifugate	0.1317
Weight Percent Centrifugate	4.4815

PLM EXAMINATION OF CENTRIFUGATE (AMPHIBOLE)			**Amphibole Asbestos Point Counts (Amphibole/Other)**					
Point Count:	Number of Occupied Points	400	Slide 1:	6	44	Slide 5:	10	40
	Number of Amphibole Asbestos Points	58	Slide 2:	8	42	Slide 6:	8	42
			Slide 3:	8	42	Slide 7:	5	45
PERCENT AMPHIBOLE ASBESTOS IN SAMPLE		**0.6498**	Slide 4:	6	44	Slide 8:	7	43
PERCENT TOTAL ASBESTOS IN SAMPLE		**1.4548**						

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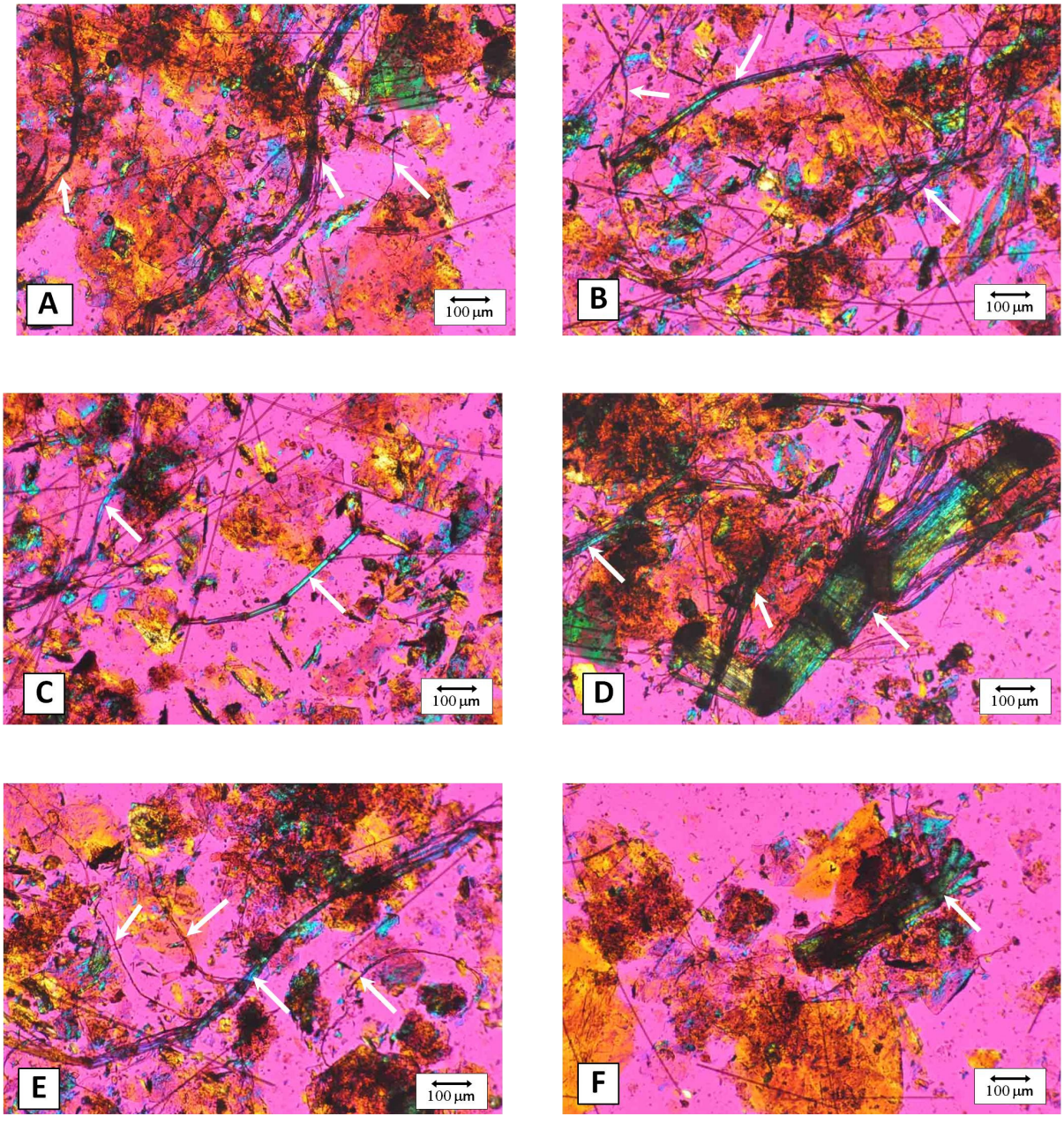
Appendix C. Examples of PLM examination of Chrysotile

Polarized light micrographs showing residue from a vermiculite-containing fireproofing sample spiked with 0.6% Chrysotile, in 1.680 refractive index liquid. White arrows indicate fibers and bundles with characteristic Chrysotile morphology.



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Appendix D. Calibration of Heavy Liquid Density

Either lithium metatungstate or sodium polytungstate can be used to prepare a heavy liquid suitable for separation of amphiboles.

As supplied, lithium metatungstate has a density of approximately 2.95 g/cc, and to separate amphiboles, the density must be adjusted to 2.75 g/cc. Sodium polytungstate may also be used, but it is supplied as a solid and a solution in water must be prepared.

Adjustment of the density is done by addition of 0.1 µm filtered water to lithium metatungstate. The density is best monitored by use of the Sink-Float® Standard. Prepare sufficient heavy liquid to complete the number of analyses to be performed.

- Place the Sink-Float® in a glass beaker, and add the required volume of heavy liquid to it, assuming that approximately 25 mL of the liquid will be used for each analysis.
- Progressively add 0.1 µm filtered water, stirring well after each addition, until the Sink-Float® begins to sink in the liquid. The ideal situation is that the Sink-Float® is suspended in the liquid, neither sinking nor floating. In general, the density adjustment is satisfactory if the Sink-Float® rises to the surface only slowly. During this process, if too much 0.1 µm filtered water is added and the density is then too low, add more of the original heavy liquid to increase the density.

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Appendix E. Required Centrifuge Times for Amphibole in Heavy Liquid

Separation of Amphibole Fragments by Centrifugation in a Heavy Liquid

The dimensions and rotation speed of the centrifuge determine the centrifugation time necessary for amphibole particles of specific sizes to sediment to the bottom of the centrifuge tubes (Herdan, 1960). The time required for sedimentation of particles can be calculated from the formula:

$$t = \left(\frac{18 \times 10^8 \times \eta}{60 \times (a - b) \times \omega^2 \times d^2} \right) \times \ln\left(\frac{R}{S}\right)$$

Where:
- t = the time in minutes for particles of diameter d to sediment
- η = the coefficient of viscosity of the heavy liquid in poise
- a = the density of the particle in g/cc
- b = the density of the heavy liquid in g/cc
- ω = the angular velocity of the centrifuge in radians/second
- d = the diameter of the particle in micrometers
- R = the outer radius of the centrifuge in cm
- S = the inner radius of the centrifuge in cm

The graph shown in Appendix E - Figure 1 were plotted for particle densities 3.0 g/cc and 3.4 g/cc using the following values:

Liquid Viscosity	=	0.09	poise
Liquid Density	=	2.75	g/cc
Angular Velocity	=	376.99	radians/second
Outer Radius of Centrifuge	=	14.5	cm
Inner Radius of Centrifuge	=	3.5	cm

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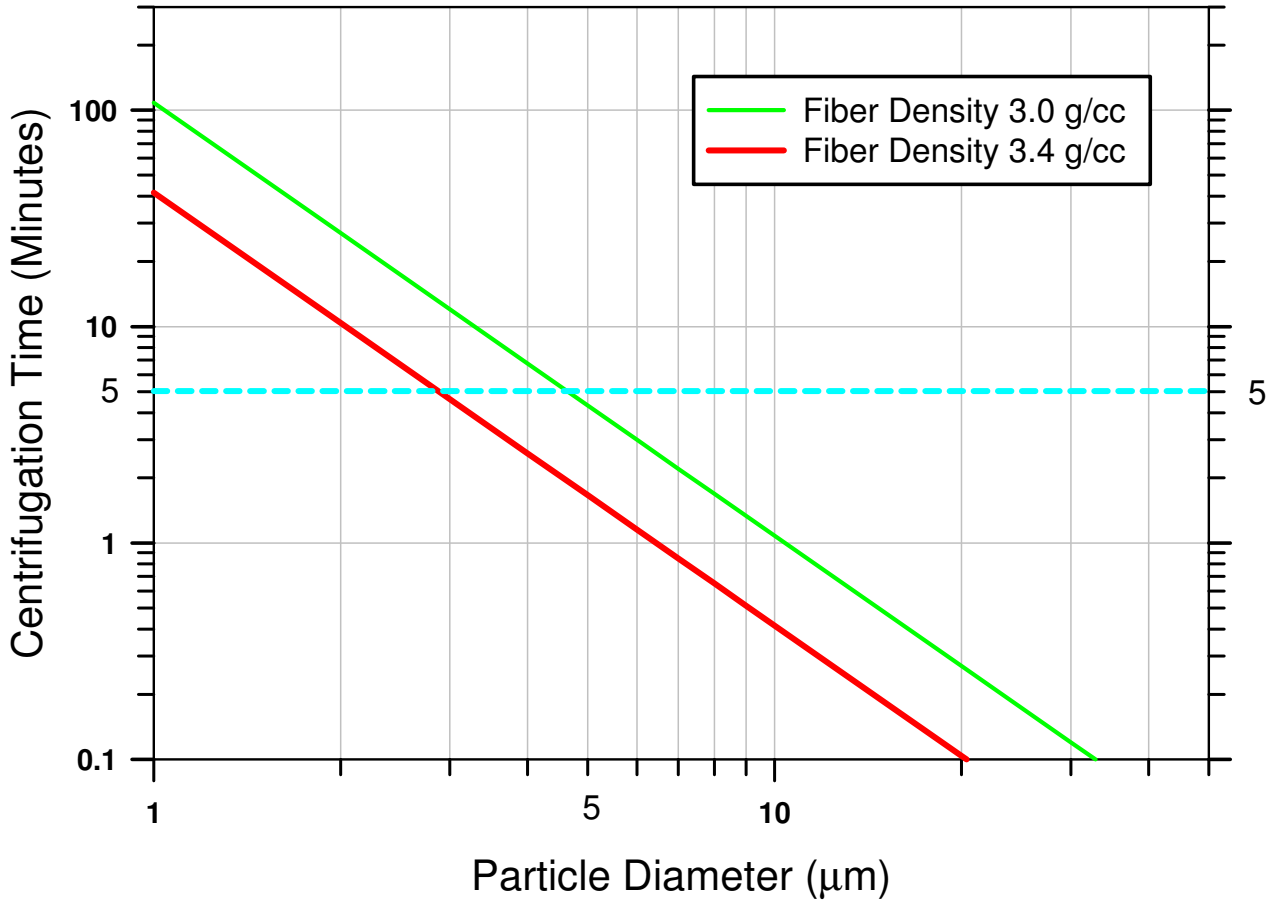
Appendix E - Figure 1: Centrifuge Times for Particles in a Liquid of Density 2.75



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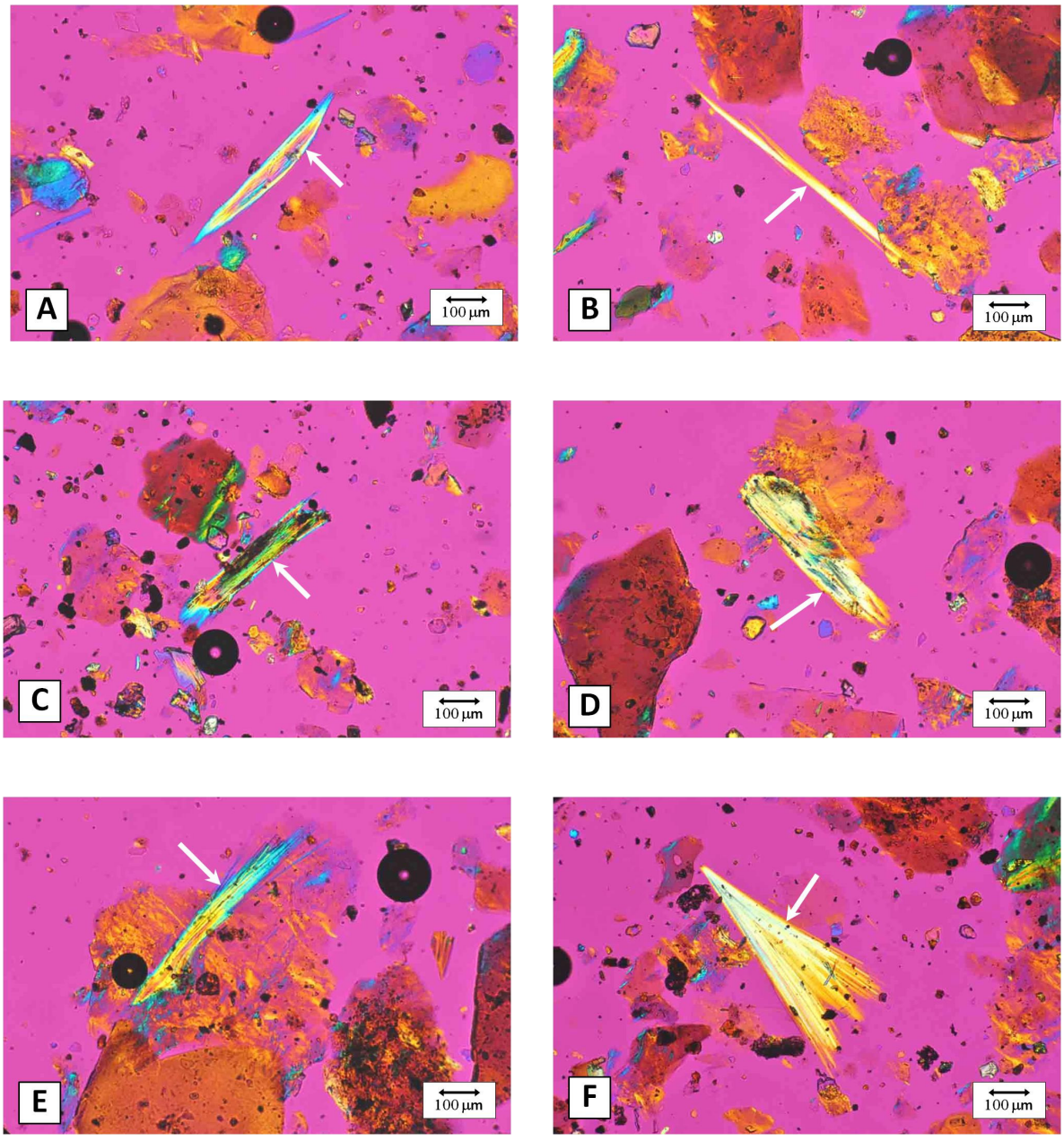
Appendix F. Examples of PLM examination of SOF-V centrifugate.

Sample contains 0.03% amphibole asbestos in 1.630 refractive index liquid. White arrow indicates fiber bundles of amphibole asbestos.



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Appendix G. Recovery of Heavy Liquid

The lithium metatungstate used in this analytical method is expensive, and it should be recovered for re-use. For the purpose of this analytical method, it is sufficient to filter it. However, after it has been used, the liquid contains a significant amount of solid material, and a 2-stage filtration is necessary. For the first filtration to remove the bulk of the solid material, use a 9 cm internal diameter Buchner funnel and a Whatman No. 40 filter, installed on a 1 liter sidearm Erlenmeyer flask. Connect a water aspirator to the flask to provide vacuum. The Whatman No. 40 filter is specified to retain particles larger than 8 µm with 98% efficiency. After filtration has completed, spray a small amount of reagent water on to the residual material collected on the Whatman filter, and re-apply the vacuum. This will assist in recovering as much of the heavy liquid as possible. Discard the Whatman filter and its contents.

The second filtration is through a 0.22 µm porosity mixed esters of cellulose (MEC) filter. This filtration is performed using a 47 mm diameter glass vacuum filtration system with a glass frit base. Although the filtration is slow, the use of this porosity of filter ensures that the filtrate meets the same suspended particle concentration criterion as that for the reagent water used in the method.

After filtration, it may be found that the recovered solution has a density lower than 2.75 g/cc, primarily because of the addition of water at the end of the first filtration. The density of this solution can be increased either by adding more of the original heavy liquid, or by heating it on a hotplate, without boiling, to evaporate some of the water. Take care to limit the amount of evaporation, so that the saturation point is not reached.

Although the recovered heavy liquids may be slightly discolored, this does not interfere with their use in this method.

Quality Assurance Qualification of Recovered Heavy Liquid

A quality assurance measurement on the recovered heavy liquid is required to confirm that it does not contain any mineral particles that would compromise analyses.

Filter 10 mL of the recovered heavy liquid through a 0.22 µm porosity, 25 mm diameter mixed esters of cellulose filter. Wash residual heavy liquid from the filter by five successive additions of 2 mL of reagent water, allowing each addition to completely filter before the next addition. Dry the filter, and prepare a microscope slide from the filter using the NIOSH 7400 preparation method. It will be found that the periphery of the filter will not clear completely because of residual lithium metatungstate. Examine the filter by PLM at a magnification of 400 to 450 using crossed polars, with a 550 nm

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(first-order red) compensator plate inserted. Examine a total of 100 fields for the presence of suspected mineral particles (birefringent particles with straight edges), excluding obvious organic materials and fibers such as cellulose. If no suspected mineral particles are detected, the liquid is satisfactory for use. If any suspected mineral particle is detected during this examination, re-filter the liquid through a 0.22 µm porosity MCE filter and test again.

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Table 2. Asbestos Types

Asbestos Types	Morphology and Color	Refractive Indices[a]		Sign of Elongation	Extinction Angle
		Perpendicular	Parallel		
Chrysotile	White to pale green. Very flexible with "kinks". Wavy with "knuckles" under PLM.	1.493-1.559	1.517-1.567	Positive	Parallel/ Undulose
Amosite	Tan. Moderately flexible but straight bundles. Easily splayed ends.	1.657-1.686	1.696-1.729	Positive	Parallel. Very infrequently shows 2° extinction.
Crocidolite	Dark blue. Flexible. Some "kinks". Splayed ends. Strongly pleochroic.	1.654-1.701	1.668-1.717	Negative	Parallel
Anthophyllite	White to light tan. Usually stiff. Ends splayed to blunt.	1.596-1.652	1.615-1.722	Positive	Parallel
Tremolite	White to light tan. Usually stiff. Large bundles may have splayed ends.	1.599-1.628	1.625-1.655	Positive	Parallel. Very thin fibers or cleavage fragments will show up to 15° extinction.
Actinolite	White to green. Usually stiff. Large bundles may have splayed ends. Often pleochroic.	1.600-1.668	1.625-1.688	Positive	Parallel. Very thin fibers or cleavage fragments will show up to 20° extinction.

[a] Perkins, R.L., and Harvey, B.W. 1993