AMA Analytical Services, Inc.
Focused On Results.

CERTIFICATE OF ANALYSIS

Chain of Custody: 300396
Client: US Food & Drug Adminitration
Address: Office of Cosmetics & Colors
4300 River Road
College Park, MD 20740
Attention: John Gasper

Job Name: Task 3 - Analysis of Official Samples
Job Location: 1st Group - 8 Samples
Job Number: CLIN 1 - Task 3 (8 Samples)
PO Number: HHSF223201810337P

Date Submitted: 3/14/2019
Date Analyzed: 3/29/2019 - 4/18/2019
Report Date: 4/25/2019
Date Sampled: Not Provided
Person Submitting: Steve Wolfgang
Revised: 4/30/2019 (1st Revision)

SUMMARY OF ASBESTOS IN TALC ANALYSIS

AMA Sample ID	Client Sample ID	TEM LOD Using ASTM D5756 Mass Calculation	TEM LOQ Using ASTM D5756 Mass Calculation	% Tremolite by TEM Using ASTM D5756 Mass Calculation	% Chrysotile by TEM Using ASTM D5756 Mass Calculation	% Total Asbestos a by TEM Using ASTM D5756 Mass Calculation	% Asbestos by PLM	% Organics	% Acid Soluable	% Other	Comments
300396-1	D-32	0.00000218%	0.00000872%	ND	ND	ND	ND	6.2%	5.0%	88.8%	
300396-1A	D-32	0.00000162%	0.00000648%	ND	ND	ND	ND	6.2%	1.4%	92.4%	
300396-1B	D-32	0.00000144%	0.00000574%	ND	ND	ND	ND	6.2%	4.0%	89.8%	
300396-2	D-33	0.00000192%	0.00000769%	ND	ND	ND	ND	28.7%	3.3%	68.0%	
300396-2A	D-33	0.00000205%	0.00000819%	ND	ND	ND	ND	29.4%	2.1%	68.5%	
300396-2B	D-33	0.00000193%	0.00000773%	ND	ND	ND	ND	29.1%	3.3%	67.6%	
300396-3	D-34	0.00000254%	0.00001016%	ND	ND	ND	ND	24.7%	4.7%	70.6%	
300396-3A	D-34	0.00000285%	0.00080274%	ND	< 0.00080%	< 0.00080%	ND	23.4%	4.5%	72.1%	
300396-3B	D-34	0.00000370%	0.00001479%	ND	0.00030%	0.00030%	ND	24.0%	3.9%	72.0%	
300396-4	D-35	0.00000134%	0.00000536%	0.00071%	0.00503%	0.00574%	ND	12.8%	12.4%	74.7%	
300396-4A	D-35	0.00000188%	0.00012905%	< 0.00013%	< 0.00013%	< 0.00013%	ND	13.7%	13.8%	72.5%	
300396-4B	D-35	0.00000168%	0.00000671%	0.00367%	0.00005%	0.00371%	ND	12.5%	15.5%	73.0%	
300396-5	D-36	0.00000188%	0.00000751%	ND	ND	ND	ND	24.0%	3.5%	72.5%	
300396-5A	D-36	0.00000114%	0.00000454%	ND	ND	ND	ND	24.1%	3.0%	72.8%	
300396-5B	D-36	0.00000150%	0.00000599%	ND	ND	ND	ND	24.1%	2.5%	73.4%	
300396-6	D-37	0.00000150%	0.00000599%	ND	ND	ND	ND	19.2%	6.7%	74.1%	
300396-6A	D-37	0.00000178%	0.00000714%	ND	ND	ND	ND	18.9%	5.3%	75.8%	
300396-6B	D-37	0.00000157%	0.00000629%	ND	ND	ND	ND	18.3%	6.0%	75.8%	
300396-7	D-38	0.00000134%	0.00000536%	ND	ND	ND	ND	0.0%	3.1%	96.8%	
300396-7A	D-38	0.00000173%	0.00000694%	ND	ND	ND	ND	0.1%	2.4%	97.5%	
300396-7B	D-38	0.00000135%	0.00000539%	ND	ND	ND	ND	0.0%	2.5%	97.4%	
300396-8	D-39	0.00000131%	0.00000524%	ND	ND	ND	ND	55.3%	11.0%	33.7%	
300396-8A	D-39	0.00000180%	0.00000721%	ND	ND	ND	ND	55.3%	8.3%	36.4%	
300396-8B	D-39	0.00000135%	0.00000540%	ND	ND	ND	ND	55.3%	15.4%	29.3%	

LOD = Limit of Detection **LOQ** = Limit of Quantification **ND** = Not Detected **PLM** = Polarized Light Microscopy **TEM** = Transmission Electron Microscopy

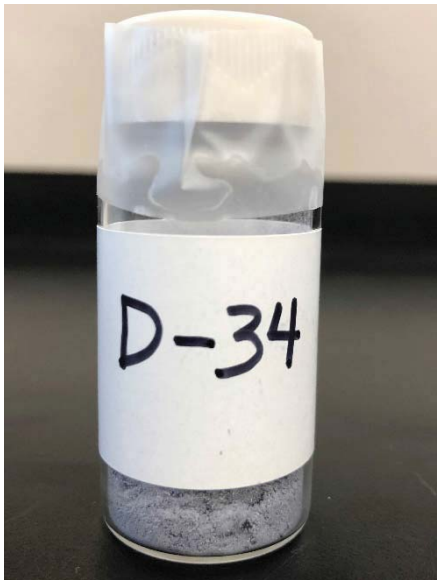
Analytical Method(s): PLM by Modified NY ELAP 198.6
TEM by Modified NY ELAP 198.4/ASTM D5756

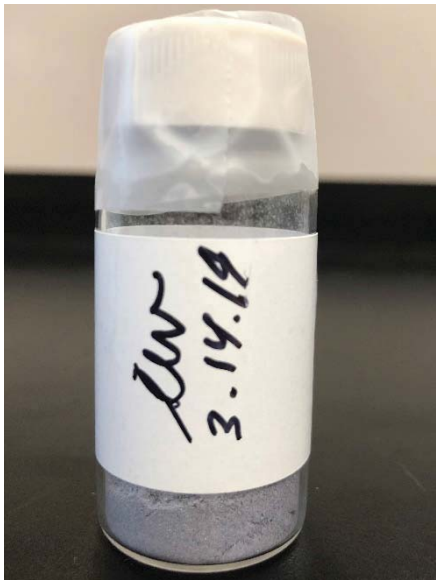
Analyst(s): **PLM**
TEM

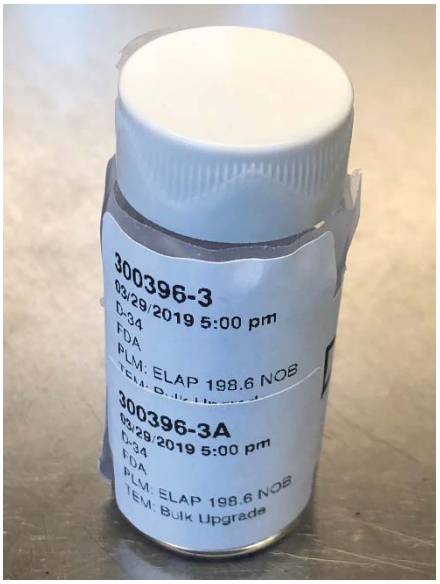
Technical Director: Andreas Saldivar

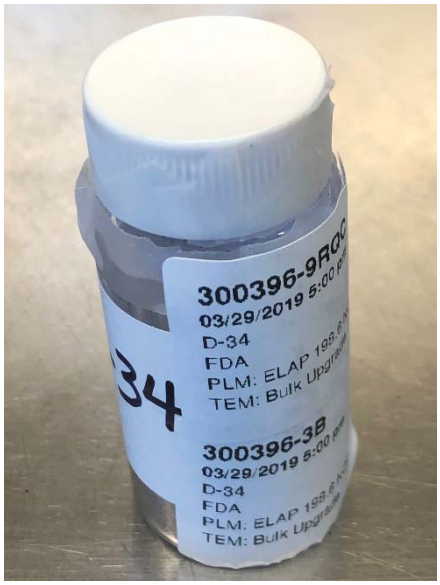
All results are to be considered preliminary and subject to change unless signed by the Technical Director or Deputy

300396-3, 3A, 3B/D-34

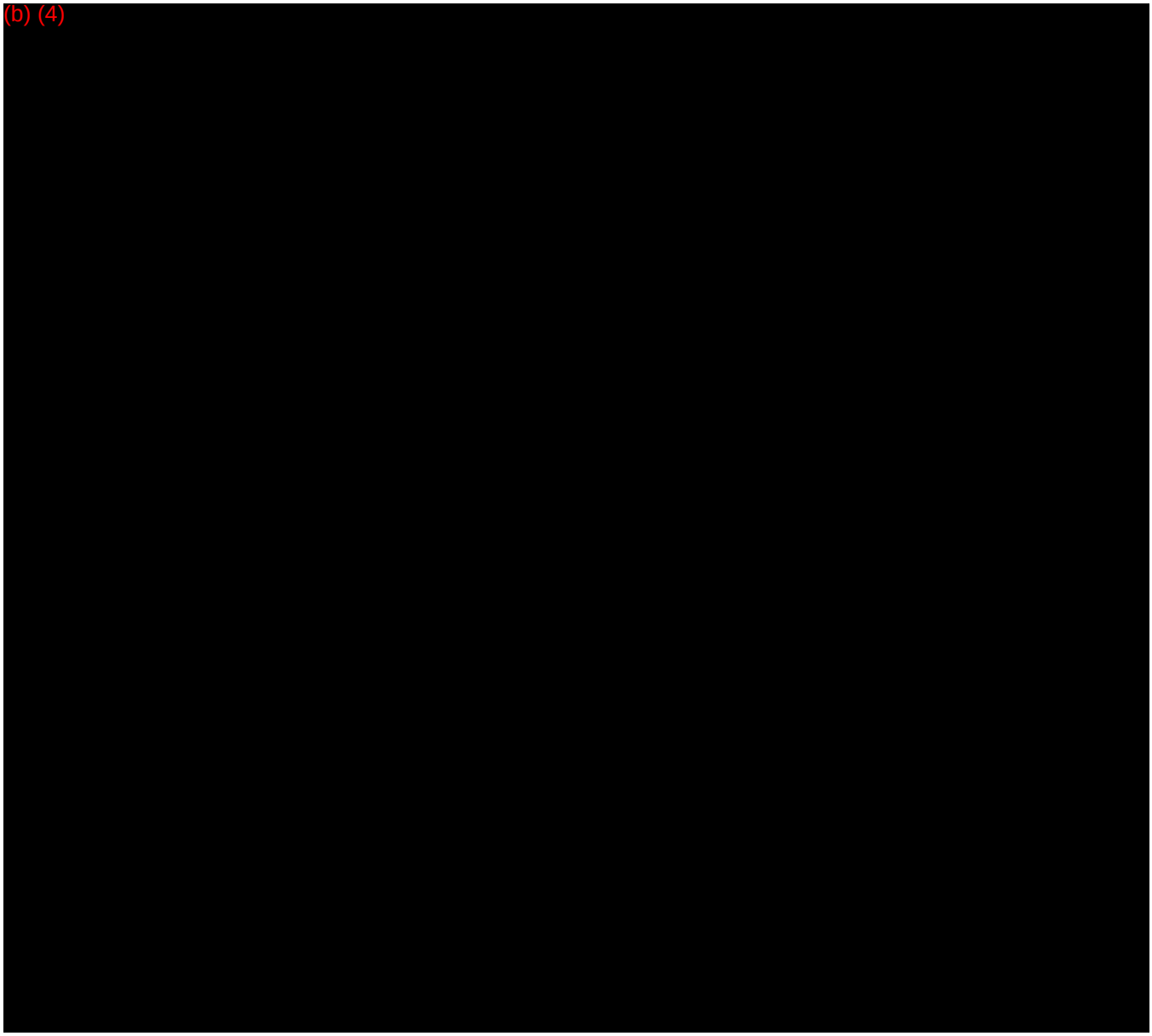








(b) (4)



Sample Preparation

Samples were prepared for PLM and TEM bulk analysis by (b) (6) on March 15, 2019 through March 29, 2019. Sample preparation consisted of the following steps:

1) Label and weigh two 8mL glass vials for each sample in the set – one vial for the PLM preparation and one vial for the TEM preparation.
2) Weigh out 0.1 to 0.8 grams of material and place in corresponding 8mL glass vial. Record weight.
3) Burn samples at 480° C for at least 12 hours.
4) Record Post-Ash Weight.
5) Treat ashed sample with concentrated hydrochloric acid.
6) Filter acid reduced material onto a pre-weighed 47mm 0.4um PolyCarbonate filter.
7) Place filter into drying oven for 30 minutes and then record Post-Acid Reduced weight.
8) Make four PLM slide preparations from the PLM residual ash for each sample in 1.550 dispersion oil. Make additional preparations in 1.605, 1.625, 1.680 and 1.700 dispersion oil as necessary for particle identification.

9) Weigh a portion of the residue from the TEM residual ash and place it into the corresponding pre-weighed 100ml jar.
10) Fill the 100ml jar with deionized water
11) Sonicate the jars for approximate 5-minutes.
12) Filter 0.2ml to 1ml of the solution onto a 47mm 0.22um MCE filter.
13) Dry the filter for 10 minutes then collapse, carbon coat, and place on a 3 TEM grids.

PLM Analysis

Analysis was performed in accordance with NY ELAP 198.6 protocols. The analysis was conducted using an Olympus BH-2 polarized light microscope (PLM) equipped with a dispersion staining objective. All four slide preparations for each aliquot were examined. 400-point count was performed for those samples on which asbestos was observed. If no asbestos was detected on any of the slides, the percentage of fibrous components was determined by visual estimation. The results of this analysis are detailed below in the *Discussion and Interpretation of Analytical Findings* section for each individual sample.

TEM Analysis

Analysis was performed in accordance with modified NY ELAP Method 198.4 protocols. The analysis was performed using a JEOL JEM-100CX II transmission electron microscope (TEM), equipped with a Thermo Fisher Quest Energy Dispersive X-Ray Analyzer (EDXA), at magnifications of 19,000x. Two grids for each aliquot were examined. Twenty (20) grid openings were examined per sample.

Modifications to the NY ELAP 198.4 Method were:

1) The residue was not placed in alcohol and prepared using the quick drop method. To obtain a more uniform preparation, the residue was placed in a jar and filled with 100ml of deionized water. The jar was sonicated, and a portion of the solution was filtered onto a 47mm 0.22um MCE filter.
2) The tremolite and chrysotile were not visually estimated. The length and width of the observed particles were measured, and the mass of each amphibole particle was calculated using the ASTM D5756 method.

The results of this analysis are detailed below in the *Discussion and Interpretation of Analytical Findings* section for each individual sample.

Calculations

ASTM D5756 Mass

$M = \pi/4\ L * W^2 * D * 10^{-12}$

M = mass
L = length
W = width
D = density

Percent Calculation

$$\frac{EFA(mm^2) * 100ml * MA(g) * RW(g)}{VF(ml) * IW(g) * AA(mm^2) * RJ(g)}$$

The calculated value is then multiplied by 100 to convert it to percent.

EFA – Effective filter area
MA – Mass of asbestos
RW – Weight of residue
VF – Volume filtered
IW – Initial weight of the sample
AA – Area analyzed
RJ – Weight of residue placed into the jar

Limit of Detection and Quantification

We used the mass of a 0.5 x 0.04-micron tremolite or chrysotile fiber, depending on what was found in each sample, as the basis for our calculations. Limit of detection was defined as 1 fiber and limit of quantification was defined as 4 fibers.

Some aliquots of samples D34 (b) (4) contained very small amounts of asbestos that were either at or below our 4-fiber limit of quantification. For these samples we defined our limit of quantification as follows:

300396-3A: mass of the single observed chrysotile fiber plus the mass of three tremolite fibers measuring 0.5 x 0.04 microns

300396-4A: mass of the two observed chrysotile fibers, the single observed tremolite structure plus the mass of one 0.5 x 0.04 microns tremolite fiber.

Discussion and Interpretation of Analytical Findings:

300396-3, 3A, 3B, Client Sample D-34

PLM

All three aliquots of sample D-34 were analyzed by (b) (6) on March 29, 2019. No asbestos or non-asbestos amphibole variants were detected the samples. The results were calculated using the equations detailed in the calculations section.

300396-3	NAD
300396-3A	NAD
300396-3B	NAD

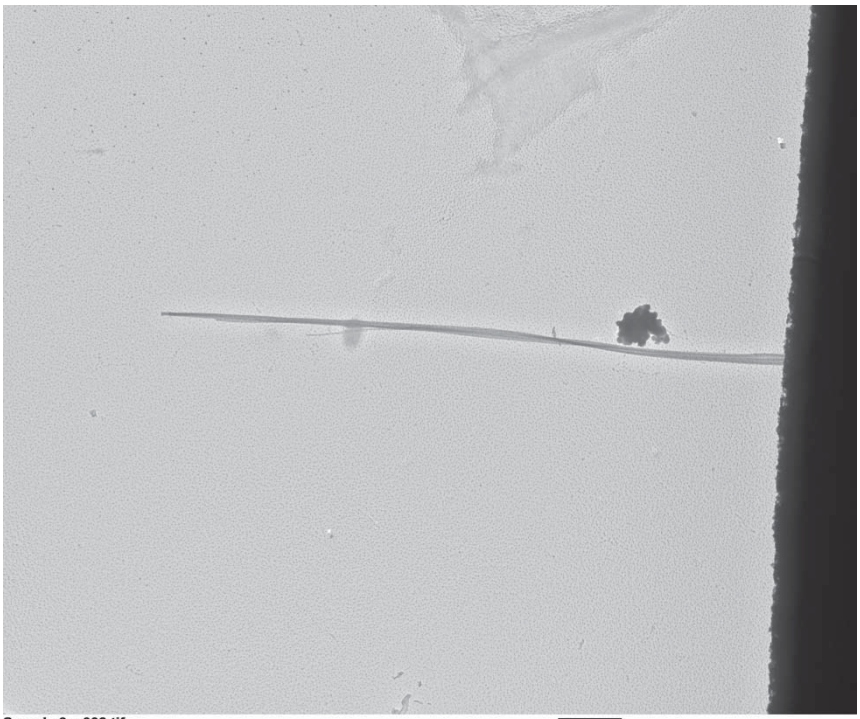
TEM

Sample 3 was analyzed by (b) (6) on April 4, 2019. Andreas Saldivar analyzed sample 3A on April 15, 2019 and sample 3B on April 16, 2019. All three samples contained mica, talc, and titanium particles. One 9.9 x 0.15 micron chrysotile bundle was counted on sample 3A. Five (5) chrysotile structures were counted on sample 3B. The results were calculated using the equations detailed in the calculations section.

300396-3	NAD
300396-3A	< 0.00080%
300396-3B	0.00030%

Below are pictures, diffraction patterns, and chemistry of the counted Chrysotile particles. The mica, talc, and titanium particles are similar to those pictured in samples 1 and 2. The unidentified peaks in chemistry spectra are copper, zinc, and carbon. Those peaks are from the TEM specimen holder and specimen grid.

Sample 300396-3A Chrysotile structure



Sample 300396-3A Diffraction pattern from the chrysotile structure pictured above.

Sample 3a_001.tif
Sample 3a Structure 1
Chrysotile Diffraction
09:58 4/18/2019
Microscopist: Andreas Saldivar
Camera: NANOSPRT5, Exposure: 800 (ms) x 5 std. frames, Gain: 1, Bin: 1
Gamma: 1.00, No Sharpening, Normal Contrast

100 (1/Å)
HV=100kV
Cam Len: 0.2200 m
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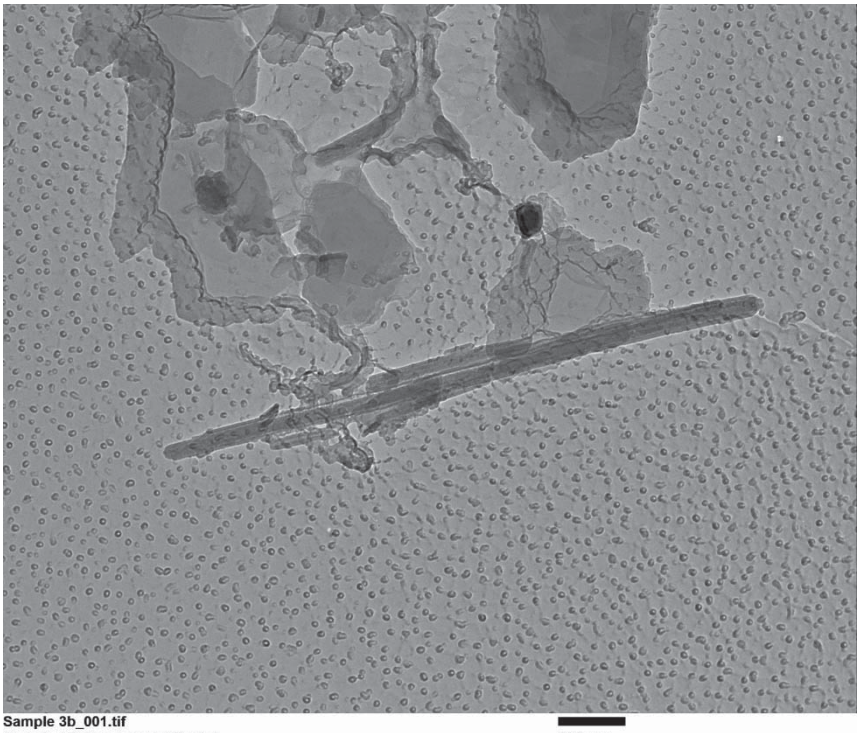
Sample 300396-3B Chrysotile structure #1



Sample 3b_001.tif
Sample 3B Structure 1 1.7 x 0.1
Chrysotile
Cal: 0.001029 µm/pix
10:16 4/18/2019
Microscopist: Andreas Saldivar
Camera: NANOSPRT5, Exposure: 800 (ms) x 5 drift frames, Gain: 1, Bin: 1
Gamma: 1.00, No Sharpening, Normal Contrast

200 nm
HV=100kV
Direct Mag: 10000 x
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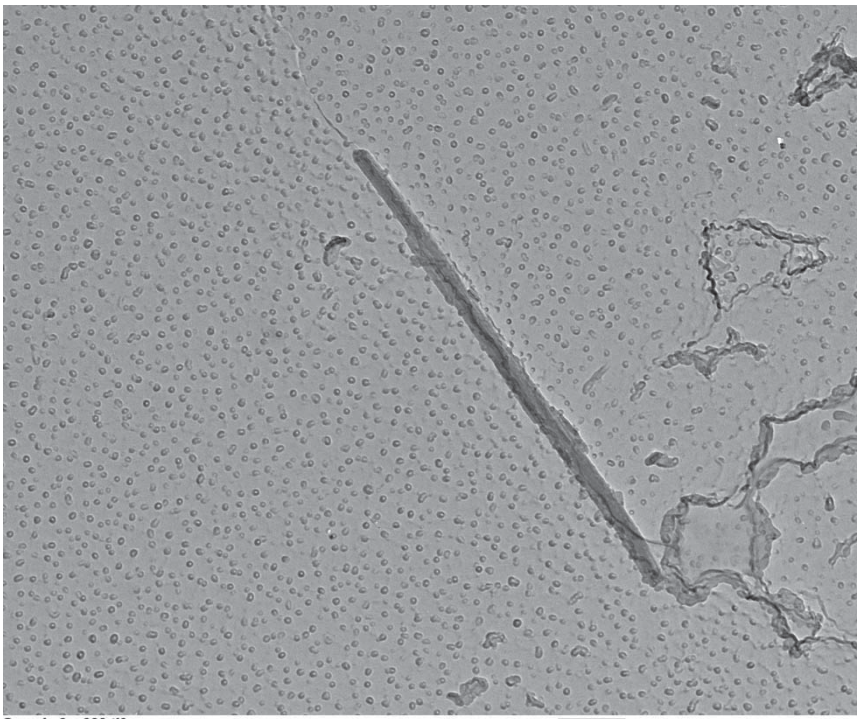
Sample 300396-3B Chrysotile structure #2



Sample 3a_003.tif
Sample 3b Structure 1.7 x 0.07
Chrysotile
Cal: 0.001029 µm/pix
10:09 4/18/2019
Microscopist: Andreas Saldivar
Camera: NANOSPRT5, Exposure: 800 (ms) x 5 std. frames, Gain: 1, Bin: 1
Gamma: 1.00, No Sharpening, Normal Contrast

200 nm
HV=100kV
Direct Mag: 10000 x
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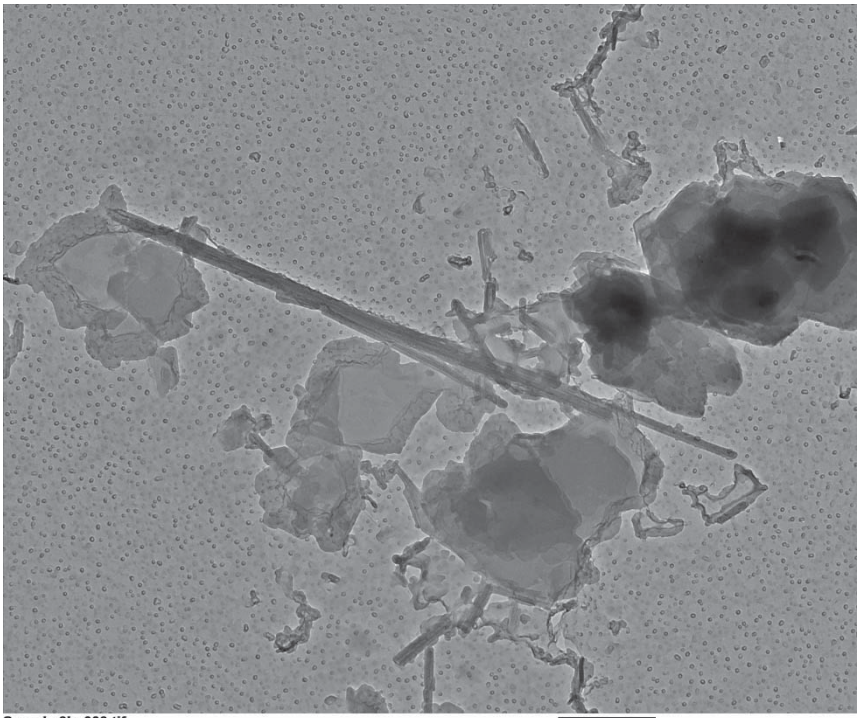
Sample 300396-3B Chrysotile structure #3



Sample 3b_003.tif
Sample 3B Structure 3 3.9 x 0.1
Chrysotile
Cal: 0.001774 µm/pix
10:24 4/18/2019
Microscopist: Andreas Saldivar
Camera: NANOSPRT5, Exposure: 800 (ms) x 5 drift frames, Gain: 1, Bin: 1
Gamma: 1.00, No Sharpening, Normal Contrast

500 nm
HV=100kV
Direct Mag: 5800 x
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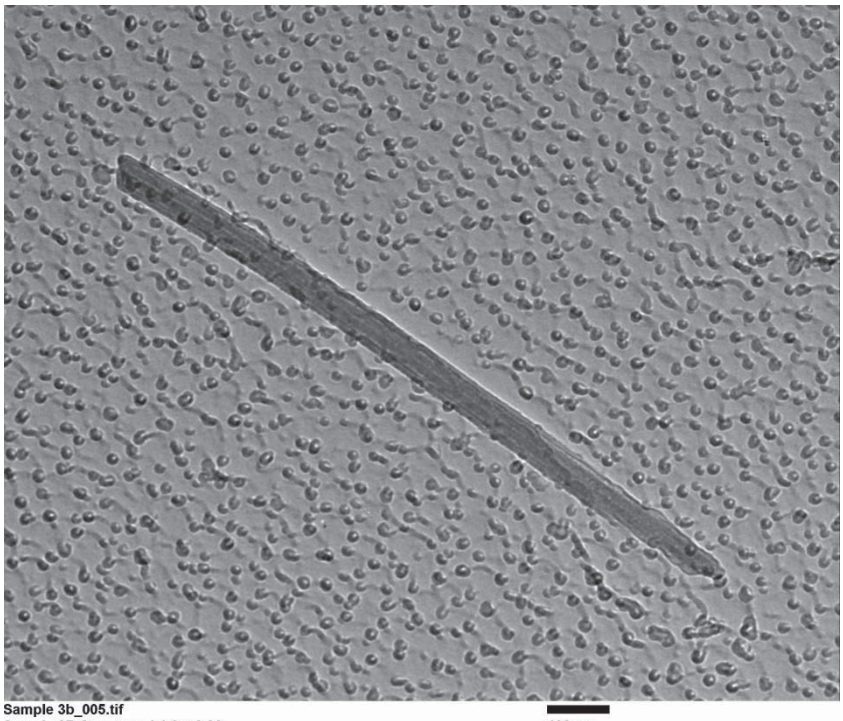
Sample 300396-3B Chrysotile diffraction pattern from structure #3

Sample 3b_002.tif
Sample 3B Structure 3 3.9 x 0.1
Chrysotile Diffraction
10:21 4/18/2019
Microscopist: Andreas Saldivar
Camera: NANOSPRT5, Exposure: 800 (ms) x 5 drift frames, Gain: 1, Bin: 1
Gamma: 1.00, No Sharpening, Normal Contrast

100 (1/Å)
HV=100kV
Cam Len: 0.2200 m
AMA Analytical Services, Inc

Sample 300396-3B Chrysotile structure #4



Sample 3b_005.tif
Sample 3B Structure 4 1.3 x 0.06
Chrysotile
Cal: 0.541520 nm/pix
10:31 4/18/2019
TEM Mode: Imaging
Microscopist: Andreas Saldivar
Camera: NANOSPRT5, Exposure: 800 (ms) x 5 drift frames, Gain: 1, Bin: 1
Gamma: 1.00, No Sharpening, Normal Contrast

100 nm
HV=100kV
Direct Mag: 19000 x
AMA Analytical Services, Inc

Sample 300396-3B Chrysotile structure #5