THE ANALYSIS OF FIRE RESIDUE PARTICLES USING OPTICAL MICROSCOPY

An Overview of Method Advantages, Limitations, and Fire-related Particle Terminology

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This document is intended as an overview of the appropriate use of microscopy methods for the analysis of fire-related particles, and the methodology used by Environmental Analysis Associates, Inc. (EAA). The document also describes our suggested color-coded data interpretation guidelines.

This document is not the laboratory Standard Operation Procedure (SOP). A copy of our SOP is available to our clients upon request. The numerical data and color-coded classifications provided in our guidelines cannot be used alone as the sole criteria to assume contamination or damage is present, or whether or not remediation is required. The laboratory analysis of submitted samples is secondary information to be incorporated into the visual inspection performed by a qualified environmental professional.

The quantitative comparison guidelines provided on page 11 are only applicable to analysis provided directly by EAA.

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1.0 OVERVIEW OF THE ADVANTAGES AND LIMITATIONS OF MICROSCOPY ANALYSIS METHODS

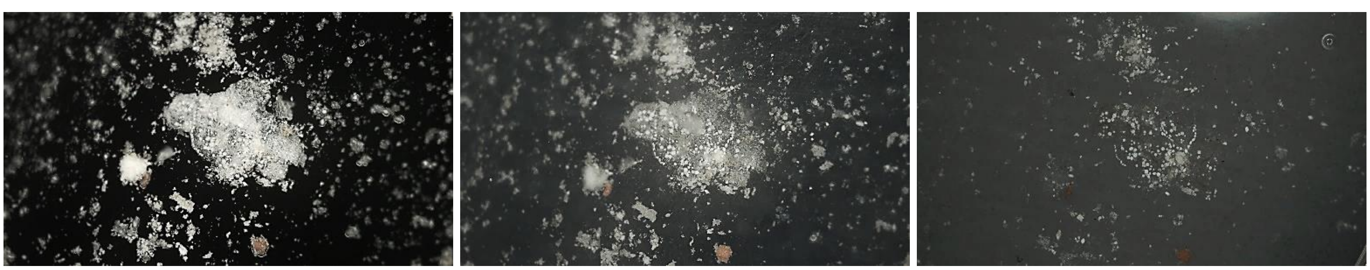
The debris generated in wildfires and structure fires are complex mixtures of burned and unburned particles. Non-resilient particles are partially burned materials that are physically fragile, semi-volatile, or semi-soluble that can be altered or destroyed by sample collection methods, laboratory sample preparation, and/or the analysis procedures.

The Advantages and Limitations of Sampling Collection and Preparation Methods

Retaining the particle morphology, chemistry, and spatial distribution during both the sample collection and analysis procedures is critically important for the proper assessment of settled combustion particles. The only sampling method capable of directly preserving the spatial relationship of dust particles found on the sampled surface is tape lift sampling. Other "indirect" methods (e.g. wipe, bulk, micro-vacuum) result in the loss of this critical information, and therefore, they are less reliable. Indirect sampling methods should only be used as secondary procedures when tape lift sampling cannot be employed.

Additionally, an unknown percentage of fire-related particles are non-resilient. The physical and chemical manipulation required to prepare the sample for analysis (e.g. sub-sampling, liquid ultrasonication, and filtration) can affect the reliability of the analysis. Wipe sampling (especially the use of wet wipes) destroys the spatial distribution patterns of the collected sample, and can breakup and dissolve ash and soot particles. This can result in the loss of particles and their identifying properties.

A dry examination of the sample (without the application of water, solvents, refractive index oils, or stains) should initially be performed. This is especially important for the assessment of ash particles generated in wildfires, and for the fragile aciniform soot clusters found as surface condensates in structure fires. The fragility and solubility of wildfire ash in liquids is of critical concern. The solubility of oak ash in distilled water is illustrated below. All micrographs were taken from a tape lift sample at a magnification of 50x using Reflected Light Darkfield Optical Microscopy.



A - "Dry" ash sample without cover slip   B - Distilled water after 20 minutes   C - Distilled water after 45 minutes

The large fire ash particle in the center of micrograph (A) was photographed dry. Distilled water was then added and a cover-slip placed on the sample and then photographed at 20 minutes elapsed time (B), and then again at 45 minutes (C). The only remaining components visible after 45 minutes in distilled water are the non-soluble oak phytolith crystals.

Aciniform soot particles can condense as large clusters or spider web-like chains on building surfaces (see micrographs on page 5), and can easily be altered through re-heating, dissolution, or disaggregation in the liquids or solvents commonly employed in some microscopic sample preparation methods. As a result, there are serious limitations in the detection and analysis of particles when using indirect sample preparation techniques (i.e. wipe sampling), and then performing the analysis by Scanning or Transmission Electron Microscopy (SEM or TEM).

The Advantages and Limitations of Optical and Electron Microscopy Analysis Methods

The fire residue analysis guidelines and methods published by the AIHA[1,2], ASTM[3], and IESO[4] have specified the use of Optical Microscopy as the primary analysis tool for the analysis of fire / combustion residues. Even with these existing published guidelines, some laboratories are continuing to recommend SEM or TEM as primary analysis methods, or as confirmation tools for the presence or absence of aciniform fire-related soot particles associated with wildfires and structure fires.

Based on our own experience with Optical and Electron Microscopy analysis methods, Optical Microscopy is the most appropriate technique for the analysis and quantification of the full size and compositional range of fire-related particles. Although Electron Microscopy can offer real theoretical resolution and elemental analysis advantages for the identification of the smaller (<10µm) resilient particle size fraction, the analysis limitations for non-resilient combustion particles (of any size) are often overlooked. Only when real-world samples are simultaneous compared using different sample preparation and analysis techniques do these limitations become obvious. The reliance of some laboratories on SEM or TEM methods as the primary analysis tool is often based on an inaccurate assumption that the ASTM D6602-13 TEM method, for the analysis of very small carbon black and other aciniform particles, will work equally well for the analysis of soot particles generated by wildfires or structure fires. Although this assumption may seem logical, it is flawed because of the inherently destructive nature of the sample preparation and analysis procedures required for Electron Microscopy. These limitations are only partially addressed in the ASTM D6602 "*Standard Practice for Sampling and Testing of Possible Carbon Black Fugitive Emissions or Other Environmental Particulate, or Both*[3]" in Sections 7.1-7.3 on pages 4-5. The appropriate use of microscopy methods is covered in more detail in the 2017 AIHA / Synergist magazine article "*The ABCs of Wildfire Residue Contamination Testing*[1]"on pages 28-29; in the 2018 AIHA "*Technical Guide for Wildfire Impact Assessments for the OEHS Professional*[2]" on pages 7-13; and in the 2010 IESO document entitled "*Evaluation of HVAC Interior Surfaces to Determine the Impact from Fire-related Particulate*[4]" on pages 14-16.

By its very design, wipe sampling, and specifically the ASTM D6602-13 TEM wipe sampling and analysis method is only capable of analyzing the resilient particle fraction that survives the sample preparation and potentially destructive TEM analysis procedures. These procedures require the dust on the wipe sampling media to be ultrasonicated to disaggregate the particles so that a liquid sub-sample can be evaporated onto the analysis media (TEM grid). A heated conductive coating is then applied under low vacuum pressure. The sample is analyzed in the TEM (or SEM) using an even lower vacuum pressure, and an image is generated using an electron beam.

The net result is an unknown amount of the fire-related particles will be significantly altered and/or lost during the sample preparation and subsequent TEM or SEM analysis.

This does not diminish the usefulness of Electron Microscopy as a tool to differentiate some of the very small aciniform (<5µm) interference particles from fire-related particles. Both the Scanning and Transmission Electron microscopes (equipped with dispersive X-ray analysis capabilities) are excellent tools for the elemental identification of small aciniform resilient particles such as metal corrosion and carbon black. Electron Microscopy is also very useful for the elemental analysis of other ash residues and identification of specific elemental cations and anions when specific preparation procedures are used to separate and analyze the soluble and insoluble fractions. This only becomes necessary when the particle properties obtained using the Optical Microscopy analysis (e.g. size, morphology, distribution, color, texture, reflectivity, crystallinity, etc.) are inconclusive, or more information is required to address specific claims related to surface corrosion or "damage". Our laboratory uses automated SEM / X-ray analysis for this specific task in the cases where it is deemed to be useful. SEM or TEM cannot be not be used as primary tools for the analysis of the wildfire or structure fire combustion by-products because neither instrument can differentiate between burned and unburned particles, and the low vacuum pressure and electron beam heating required by the analysis will result in the loss of the non-resilient fire-related particles.

Although Optical Microscopy techniques have a more limited magnification range for the analysis of very small particles (when compared to Electron Microscopy), the broad range of simultaneous and non-destructive optical measurement properties available provide the best balance of advantages, and the least number of limitations in providing a reliable analysis.

2.0 THE EAA OPTICAL MICROSCOPY ANALYSIS METHOD FOR WILDFIRES AND STRUCTURE FIRES

The EAA fire / combustion residue analysis method is preferably performed on tape lift samples so that the full range of Reflected Light and Transmitted Light Microscopy can be performed. A properly equipped optical microscope is used to employ Reflected Light Darkfield, Transmitted Light Bright Field, and Polarized Light Microscopy simultaneously. The EAA methodology is described in our laboratory Standard Operating Procedure (SOP).

The EAA method first provides an initial qualitative dry examination of the color, texture, and presence or absence of large soot, char, or ash particles found on the entire tape lift slide preparation. The qualitative examination is followed by a quantitative estimate of the fire / combustion residue particles found in the sample. The adhesive tape lift sample analysis procedure preserves the integrity, size and spatial distribution of the deposited dust, and is used to simultaneously estimate the area ratio percentage and the concentration of settled fire-related particles on sampled surfaces per unit area (cts/mm^2). The analysis of bulk dust and micro-vacuum samples can also be used to estimate the area ratio percentage of fire-related particles within the sample, and to determine other chemical properties such as pH, conductivity, and corrosion potential. These other EAA analysis procedures are covered in a separate document.

Wild fire / vegetation fire related particle distributions

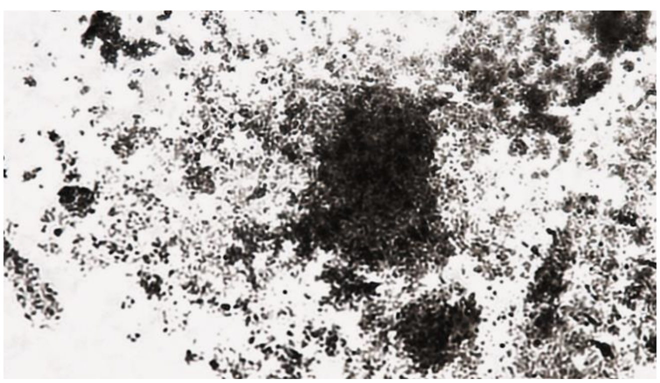
The background of fire / combustion particles (soot, char, & ash from vegetation and anthropogenic sources) measured in non-impacted commercial office and residential spaces are typically below 1% (area ratio%) with a range of 0.1% to 3%. In some micro-climatic, geographic, or industrial locations, isolated measurements as high as 5% may occur. This inherent background of outdoor and indoor sources includes but is not limited to, controlled burning, vehicular emissions, asphaltic materials from roadways, wood burning fireplaces, candles, cooking, industrial activity, fossil fuel power plants, and re-entrainment of dust from historical wildfires.

Wildfire or burned vegetation particles, when found inside structures, are primarily composed of char and ash particles and not aciniform soot. The lighter aciniform soot particles generated from the volatile plant resins are typical lofted high into the air in the crown of the fire, and are not routinely found as condensed or settled debris inside structures unless the structure is within the burn zone. As stated above, the char and ash particles can be extremely fragile and the ash particles can contain high concentrations of soluble material.

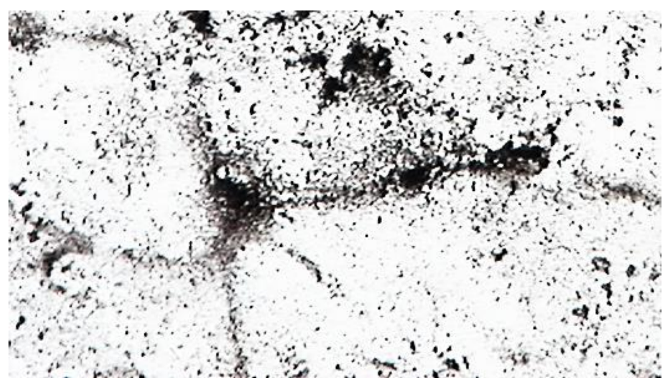
The char and ash particles from large wildfires can often be detected downwind over 50 miles away. Indicator particles associated with specific types of fire events (e.g. identifiable leaf, twig, and bark char; burned pollen, plant parts, trichomes, and phytoliths) are particles that can be associated with specific vegetation burning, or wildfires. Intact indicators of burned vegetation are typically found in significantly lower concentrations than the general population of vegetative char and ash particle residues, especially as the distance from the fire increases. Other non-vegetative wildfire indicator particles associated with firestorm winds can include burned soil / clays and carbonized quartz grains. The EAA laboratory analysis provides quantitative estimates of these indicator particles as part of the fire-related debris in the sample.

Structure fire related particle distributions

Unlike wildfires, structure fires generate an entirely different distribution of soot, char, ash, and indicator particles. Because the heated plume of a structure fire is usually contained within the building envelope, the fuel based and heated organic compounds form aciniform soot condensates that can deposit as larger clusters (e.g. 50-100μm), or even larger spider web-like chains on cooler ceiling and wall surfaces within the structure. The depositional patterns generated by aciniform soot condensation are critical diagnostic indicators for an indoor fire, and can be readily observed using an Optical Microscopy examination of tape lift samples collected directly from walls, ceilings, or contents (see the example micrographs below).



Structure fire soot aggregate clusters - 600x



Structure fire soot spider web-like chains - 600x

These diagnostic in situ depositional patterns are not preserved on wipe samples or other indirect sampling methods.

The larger char and ash particles generated during a structure fire end up settling and depositing on horizontal surfaces. The composition of char, ash, and oxidation particle residues is determined by what has actually burned (paper products, wood framing, plastics, metals, and/or proteinaceous materials, etc.). Additionally, the background levels found in industrial and warehouse spaces can vary widely depending the activities and potential fuel sources in these locations. Control samples may be required to differentiate Typical background levels from Atypical or Elevated conditions in these locations. As a result, the color-coded guidelines provided on page 11 may not directly apply to these environments. Other methods such as SEM analysis, pH, conductivity, and/or cation/anion analysis may also be required to fully assess these complex types of structure fires. The SEM analysis can also be performed directly from the same tape lift samples thereby preserving the original integrity of the settled particles.

3.0 FIRE COMBUSTION TERMINOLOGY AND BASIC DEFINITIONS

The following four major combustion particle classifications are used by EAA in the laboratory reports:

- **Aciniform soot -** Differentiated as fine or large clusters where possible
- **Char** – Differentiated as vegetative and/or non-vegetative where possible
- **Ash** – Differentiated as vegetative and/or structure-related where possible
- **Indicator / assemblage particles** – Differentiated based on origin or source for both wildfire and structure fires

The definitions for the industry recognized wildfire combustion particle generic categories (soot, char, and ash) are illustrated in Figure 5 on page 10 of the April, 2018 AIHA "*Technical Guide For Wildfire Impact Assessments. A Guide for the Occupational and Environmental Health and Safety Professional*", and shown below.

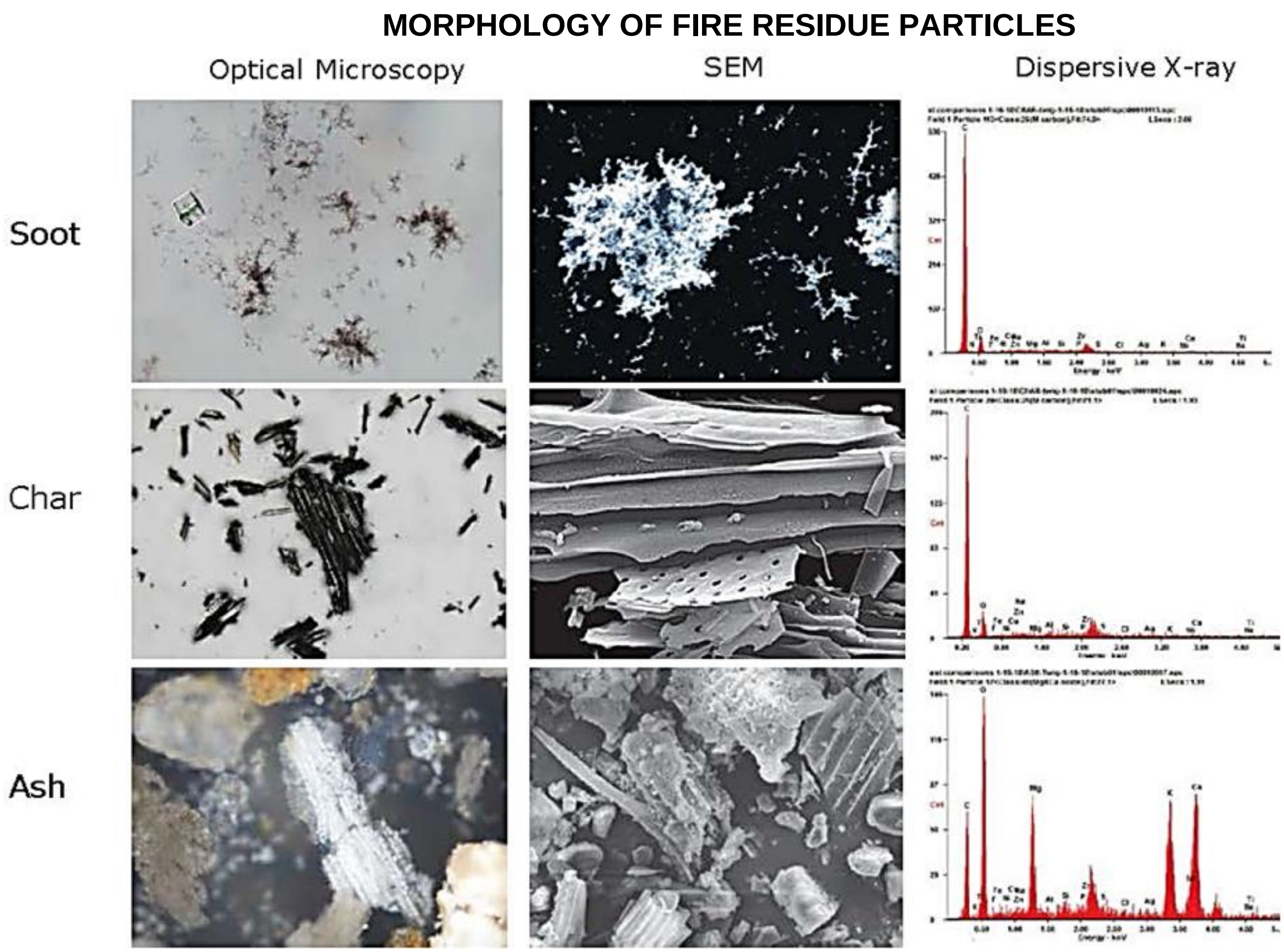
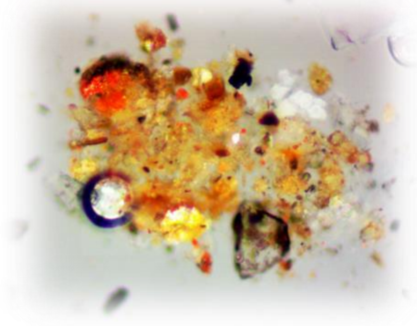


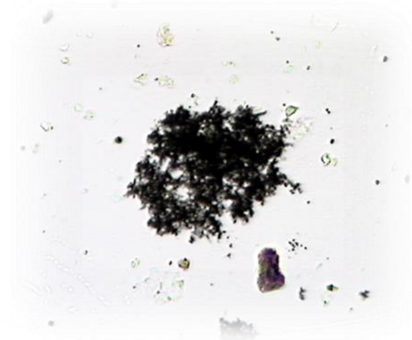
Figure 5. Technical Guide For Wildfire Impact Assessments. A Guide for the Occupational and Environmental Health and Safety Professional.

Example EAA micrographs of the soot, char, ash, and associated indicator particles found in both wildfires and structure fires are illustrated below.

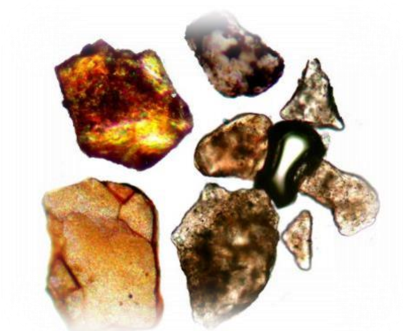
EXAMPLE WILDFIRE / VEGETATION COMBUSTION PARTICLES

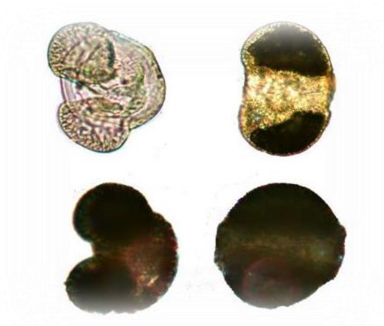

Char

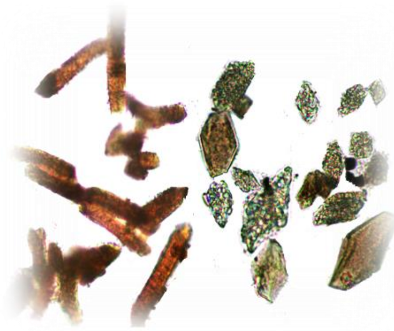

Ash (with Phos-chek fire retardant)


Aciniform Soot (rare)

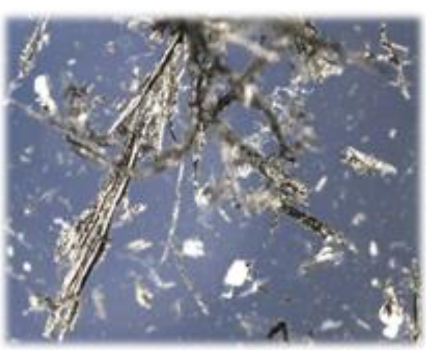
EXAMPLE INDICATOR PARTICLES

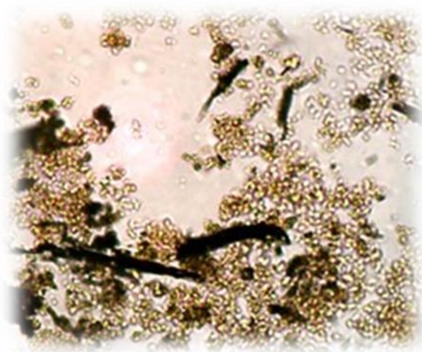

Burned soil, quartz, clays


Pollen **-** Normal → singed → burned


Plant phytoliths

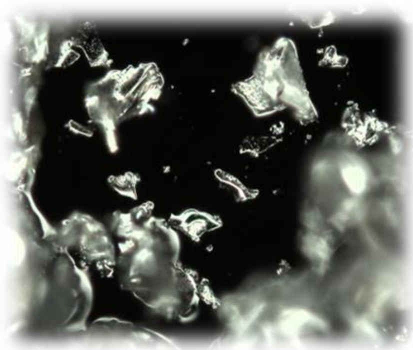
EXAMPLE STRUCTURE FIRE PARTICLES

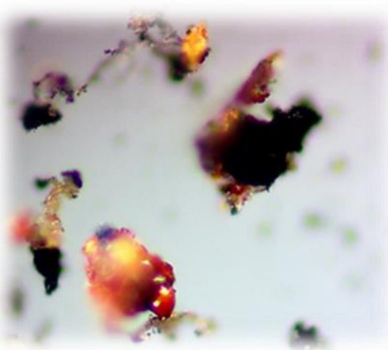

Char – cardboard

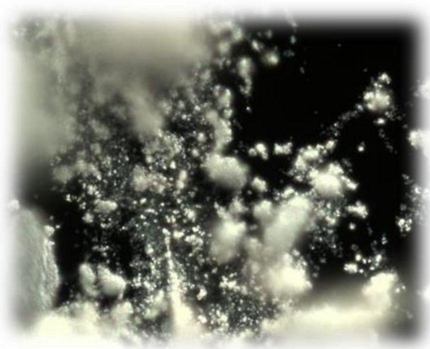

Mixed char / ash – copier paper


Aciniform Soot (Common)

EXAMPLE INDICATOR PARTICLES


Melted PVC plastic


Melted wire insulation


Burned paint residue

The definitions for each fire-related particle classification are described below:

a. **Aciniform soot -** Black/brown carbonaceous particle clusters that have a chained grape-like structure that are most often the result of organic or fossil fuel combustion. Soot clusters associated with wildfires are not typically found indoors unless the plume is impinging directly on the structure. Large soot aggregates (e.g. ~50-100µm) are potential indicator particles found in structure fires as condensates on ceiling and wall surfaces. These larger aciniform soot clusters or chains (when found in significant concentrations indoors) are rarely the result of exterior or anthropogenic infiltration sources. The diagnostic soot aggregate depositional patterns characteristic of a fire event can only be observed by using tape-lift samples to collect the unaltered particles directly from the impacted surface. This diagnostic property is lost when the samples are collected and subjected to disaggregation. The individual disaggregated particles within the aciniform cluster are usually too small to accurately identify or quantify using Optical Microscopy due to their small size (i.e. <1µm).

b. **Vegetation Ash –** Silvery gray to white soluble mineral salts (primarily Calcium, Potassium, Magnesium oxides/hydroxides) and insoluble plant phytoliths remaining after combustion and removal of carbonaceous material from burned vegetation (wildfires). Fresh ash can have elevated pH levels of 10-13. The optical properties of the potentially soluble ash constituents can only be observed by using tape-lift samples to collect unaltered particles directly from the impacted surface. These soluble materials can only reliably be observed by first performing a dry examination in reflected light darkfield optical microscopy. The non-soluble residues contain high concentrations of calcium oxides / carbonates and siliceous plant phytoliths.

c. **Structure fire Ash –** Residual minerals, chlorides, decarbonized paint, and plastic residues remaining from the burned contents. The particulate residues are classified in the indicator particle categories when they can be identified. The soluble components in these residues are typically composed of chloride residues with pH values ranging from approximately (3-6).

d. **Char – Vegetation / Wood -** Pyrolyzed (burned) vegetation with visual evidence of the plant cell structure. Vegetation char particles generated in wildfires is typically represented by the parts of leaves, stems, and bark.

e. **Non-vegetation Char -** Pyrolyzed (burned) angular brown / black particles showing no surface structure or inconclusive residual morphological evidence of combusted cellulose or a woody plant structure. Non-vegetation char can also include vegetation derived paper products where the cellulose fibers have been altered and can be observed to be overlaying each other in a cross-wise fashion.

f. **Assemblage Indicator Particles –** Burned or singed particles with a known source, or morphological or chemical properties that are characteristic of a specific fire source or origin. These particle types are described below.

   1. **Vegetation / wildfire indicator particles – Indicator particles generated from vegetation burning or the wildfire environment**

      - **Plant Phytoliths –** The residual and rigid, microscopic crystal structures made of silica or calcium oxalate, found in plant tissues that are resilient and remain after the decay or combustion of the plant. These particles are associated with the fully decarbonized residual crystalline ash components of wildland or vegetation fires. Sometimes the vegetation source or origin of these materials can be determined. A full assessment of the vegetation origin may require additional samples and analyses.

- **Burned clays / quartz or mineral grains –** Soil minerals (clays, quartz, etc.) that exhibit heating during intense fires. Clear quartz or other translucent minerals with a carbonized coating are an indicator of wildfire exposure often associated with firestorm winds.

- **Burned pollen grains & plant trichomes –** Pollen grains and trichomes (leaf hair fragments) and other plant structures that have been singed or burned.

- **Fire retardant particles –** Pink / orange chemicals typical of Ammonium phosphate such as Phos-Chek used in aircraft drops to inhibit the spread of wildfires.

2. **Structure / protein fire indicator particles – Indicator particles generated from the combustion of structural construction materials, contents, or protein burning**

- Aciniform soot – Clusters (3-100µm) or web-like chains of black/gray aciniform particle deposition in transmitted light, and black-silver in Reflected Light Darkfield illumination. These optical properties can only be observed by using tape-lift samples to collect in situ unaltered particles directly from the impacted surface.

- Droplet-like opaque particles from melted or condensing materials that can be found in grape-like clusters or a wide range of dark droplet-like particles. These optical properties can only be observed by using tape-lift samples to collect unaltered particles directly from the impacted surface.

- Burned construction materials - Construction materials (mastics, asphalt, drywall, plasters, shingles, OSB, Douglas fir lumber) that exhibits partial combustion / heating during intense fires.

- Combusted paint particles – Primarily residual Titanium dioxide with minor amounts of chlorides, iron, or other pigments.

- Oxidation / corrosion particles (Aluminum, iron, zinc, oxides)

- Melted Plastics – Residual PVC, chlorides, polycarbonates, rubber, wire insulation, etc.

- Singed, melted, or burned cellulose and fabric fibers.

- Smooth and semi-soluble grease-like particles from the burning of proteinaceous material.

g. **Interference particles –** The use of the term interferences is used in our fire residue analysis report to qualitatively indicate the presence of a surface dust condition where the physical, chemical, morphological, or depositional density (masking) properties of the dust distribution interfere with the analyst’s ability to resolve, identify, or differentiate specific particles within the sample. This condition may result in the lowering of the analysis accuracy and precision. The reporting of interferences does not necessarily affect the identification of specific fire residue particles unless specifically indicated.

h. **Opaque particles –** Refers to the optical properties of a particle in transmitted light. Opaque particles in general can appear black or brown (in transmitted light) or simply indicate the absence of significant light transmittance. There are a large number of biogenic and anthropogenic (man-made) sources that are not combustion related and are classified and placed in this category within the EAA laboratory report. These particles include tire rubber, biogenic decayed debris, soil particles, paint, corrosion, etc.

i. **Area Ratio % -** The estimated total projection area of each individual particle category (e.g. soot, char, ash) as compared to the total area of all other particles in the sample.

j. **Surface Particle Density (cts/mm^2) –** The estimated numerical surface concentration per unit area (cts/mm^2) of each particle category. The surface density can only be measured on tape lift samples.

4.0 DATA INTERPRETATION

The EAA analysis report provides both qualitative and quantitative parameters to determine if the measured fire / combustion residue concentrations for a specific site are Typical, Atypical, Elevated above background, or associated with a specific fire event. The following considerations need to be followed when interpreting the EAA report:

1. The laboratory data, and summary guidance tables cannot be used by themselves as a direct indicator of contamination, impact, or the need for remediation. The reported results (and classification ranges shown below on the following page) apply to typical commercial and residential buildings. The quantitative results are secondary information to be integrated into the visual inspection and site-specific conditions documented by a qualified environmental professional.

2. Qualitative observations - Qualitative observations of odor, color, texture, and the presence or absence of large fire combustion particles (aciniform soot clusters, char, ash) are important indicators of the proximity or recent history of the deposited material.

3. Quantitative measurements - The reporting of area ratio percentages and the surface particle deposition (particles/mm^2) are used to determine the magnitude of surface fire-related debris concentrations. The presence or absence, and concentration of indicator or assemblage particles are also quantified to assist in the determination of the potential source or sources of the fire. There are advantages and limitations to each of these assessment techniques and methods, and each method used by itself may not fully characterize the sample. In other words, there is no singular analytical metric that can be used to assess the fire-related impact all by itself.

4. Data variability - As a part of comparing quantitative estimates, it is important for the investigator to consider both the inherent sampling variability (even between samples collected from the same location), and the analysis variability. EAA tracks the microscopic variability (CV) in our laboratory.

EAA provides a basic comparison chart dividing the quantitative data into four color-coded concentration ranges: *Typical*, *Typical-upper background*, *Atypical*, *and Elevated*. These ranges are based on the total enumerated concentrations (area ratio %) and surface density (cts/mm^2) of fire-related particles, and take into account the background from other potential sources. These ranges should be used for preliminary comparative estimates only. (Determining if *Atypical* or *Elevated* measurements are related to a specific fire event must be based on all information gathered during the site investigation and not rely on the laboratory data alone). The suggested color-coded concentration classification ranges are described on the following page.

The concentration levels do not automatically imply contamination, damage, a health impact, or the need for remedial action. The ranges are also based on their multiplicative difference from database background or control samples (e.g. 3 or 10 times the background control range). It is also suggested that where-ever possible, onsite controls specific to the individual project conditions should also be used for comparisons with background and assessing impact.

a. **Typical-low -** Normal background concentrations (the approximate 50th percentile range) found in non-impacted commercial and residential buildings.

b. **Typical–upper background** – Upper background range accounting for typical indoor or infiltration sources from cooking, candle burning, fireplaces, barbecues, roadway sources, etc. This upper range also takes into account the inherent sampling and laboratory analysis variability.

c. **Atypical** – Concentrations measured approximately 3-10 times above the normal background range potentially associated with singular or multiple fire sources. It cannot be assumed that concentrations measured in this range are associated with a specific event without additional information regarding the site-specific conditions, the presence of indicator particles and the presence or absence of potential contributing sources.

d. **Elevated** – Concentrations measured approximately10 times the normal background range. Concentrations in this range are most likely associated with a specific fire event or chronic exposure to multiple events. Levels in this range still need to be evaluated in the context of the site-specific conditions, the presence or absence of specific indicator particles, and the presence or absence of potential contributing sources.

Total Area Ratio % and Numerical Surface Concentrations		
Classification Range	Total Fire Residue (ratio%)	Total Fire Residue (cts/mm^2)
Elevated > 10x background	>10%	>50
Atypical 3 -10x background	3-10%	5-50
Typical - upper background	1-3%	1-5
Typical - low	<1%	<1

The measured ratio % and the surface density (cts/mm^2) ranges are impacted differently based on the particle size distribution and actual amount of background dust found on the sampled surface. For this reason, qualitative observations, quantitative measurements, and the presence or absence of indicator particles all need to be considered in the final interpretation of the laboratory results.

5.0 REFERENCES

1. Daniel Baxter, Russ Crutcher, Brad Kovar, Larry Wayne, August, 2022. Analysis of Wildfire and Structure Fire Combustion Residues, Microscopy and Other Considerations, The Synergist Magazine.

2. Daniel Baxter, Alice Delia, Susan Evans, Brad Kovar. et.al, April, 2018. AIHA Technical Guide For Wildfire Impact Assessments. A Guide for the Occupational and Environmental Health and Safety Professional. American Industrial Hygiene Association.

3. ASTM D6602–13, reapproved 2018. Standard Practice for Sampling and Testing of Possible Carbon Black Fugitive Emissions or Other Environmental Particulate, or Both.

4. Alice Delia & Daniel Baxter, November 2017. The ABC's of Wildfire Residue Contamination Testing – Post Fire Assessments of the Indoor Environment, The Synergist Magazine.

5. IESO / RIA Standard 6001, 2010. Evaluation of Heating, Ventilation and Air Conditioning (HVAC) Interior Surfaces to Determine the Presence of Fire-Related Particulate as a Result of a Fire in a Structure.