

TENNESSEE BUREAU OF INVESTIGATION

Forensic Services Division

Microanalysis Standard Operating Procedures Manual

Gunshot Primer Residue Analysis



Analysis for Gunshot Primer Residue Using Scanning Electron Microscopy/Energy Dispersive Spectroscopy (SEM/EDS)

1. Scope

This method describes the analysis of micrometer size particles to determine if they contain any combination of antimony, barium, and lead. These elements are the main components of gunshot primer residue (GSR). GSR can be deposited on items and subjects when a firearm is discharged. This analysis is performed using a Scanning Electron Microscope with Energy Dispersive Spectroscopy (SEM/EDS).

2. Terms and Definitions

Scanning electron microscope (SEM) – A form of microscopy using electrons rather than visible light to obtain high resolution images over a dynamic range of magnification.

Energy Dispersive X-ray Spectrometry (EDS) –Makes use of the x-ray spectrum emitted by a solid sample bombarded with a focused beam of electrons to obtain a localized chemical analysis.

Sample stub – a specimen holder/collection device for examination using SEM/EDS usually made of aluminum. The stub consists of a wide, flat sample surface and a pin for mounting on the SEM stage.

Adhesive tab – round, carbon-based, double sided adhesive disc designed to be placed on a sample stub for GSR evidence collection.

Carbon tape – carbon-based, double sided adhesive tape that may be cut into short sections and placed on a sample stub for GSR evidence collection.

Peak overlaps - There are energy peak overlaps among different elements, particularly those corresponding to x-rays generated by emission from different energy-level shells (K, L and M) in different elements. Particularly at higher energies, individual peaks may correspond to several different elements.

3. References

Guide for Primer Gunshot Residue Analysis by Scanning Electron Microscopy/Energy Dispersive X-Ray Spectrometry, SWGGSR

TENNESSEE BUREAU OF INVESTIGATION

Forensic Services Division

Microanalysis Standard Operating Procedures Manual

Gunshot Primer Residue Analysis



ASTM E1588 - Standard Practice for Gunshot Residue Analysis by Scanning Electron Microscopy/Energy Dispersive X-Ray Spectrometry

INCA Feature Instruction Manual, Oxford Instruments, Chapters 2 & 3.

The Principles and Practice of X-ray Microanalysis (compact disc) by Oxford Instruments

Aerospace Report No. ATR-77 (7915)-3 Equipment Systems Improvement Program, Final Report on Particle Analysis for Gunshot Residue Detection, Law Enforcement Development Group, Aerospace Corporation, September 1977.

4. Examination Procedures

4.1. Evidence Types

Commercially supplied SEM/EDS gunshot primer residue kits for subject hands are the primary evidence type for this analysis. Other evidence for GSR SEM/EDS analysis may include, but is not limited to: the outer clothing of the subject(s), vehicle(s), and items in the area of a firearm discharge. Generally, hand kits and clothing from an individual that has been wounded by a gunshot will not be processed. This individual has already been associated with a firearm discharge.

4.2. Reagents and Chemicals

4.3. Procedural and Chemical Precautions

Refer to the TBI Safety Manual for general safety requirements.

When carbon coating samples, the carbon rods should only be observed using goggles rated for welders and cutters. The goggles conform to ANSIZ87.1.

When filling a dewer flask with liquid nitrogen to be used in the Scanning Electron Microscope/Energy Dispersive System (SEM/EDS), protective clothing must be worn. This includes cryogenic gloves, full-face shield and laboratory coat.

Protective attire, including laboratory coat, mask, gloves and eye protection should be used when working with clothing and/or bloodstained items.



TENNESSEE BUREAU OF INVESTIGATION

Forensic Services Division

Microanalysis Standard Operating Procedures Manual

Gunshot Primer Residue Analysis

Decontamination of a scientist's work area should be performed after each use, but must be done after analyzing bloodstained items.

4.4. *Quality Assurance Procedures*

1. A positive standard for particles of gunshot primer residue will be created by firing ammunition with standard composition primers. A sample stub with a sticky tab will be used to collect particles of GSR. This stub will be analyzed to ensure that gunshot residue particles one (1) micrometer and larger have been collected. Some particles smaller than one (1) micrometer may also be detected. All these particles shall contain antimony, barium and lead.

This positive standard stub will be analyzed repeatedly and a distribution map of the particles will be made to confirm reproducibility. Once this step is completed, this stub is the positive control for the SEM and will not be moved or removed from the SEM stage until it is necessary to create a new positive control.

2. Regular monitoring of the microanalysis laboratory environment can determine if the precautions taken and cleaning routines are effective. Suitable areas to be monitored include GSR sample collection areas, the SEM/EDS area, and exterior surfaces of the carbon coater. The monitoring of these areas shall be performed semi-annually. These results will be maintained in the Environmental Monitoring notebook. The stubs will not be kept.
3. Negative control samples are analyzed with each batch of stub samples. These control samples monitor the sample coating preparation and the SEM interior chamber.
4. If contamination is indicated in the negative control sample:
 - a. The incident shall be diagnosed to determine the exact source.
 - b. Case impact shall be assessed.
 - c. The areas indicated by the contamination should be thoroughly cleaned and tested for residual contamination.
 - d. Case work can resume once the contamination procedures have been demonstrated to have been effective.
 - e. A report may be issued stating the negative control was contaminated and no analysis was performed.
 - f. If a systemic problem is found, a corrective action will be applied.



TENNESSEE BUREAU OF INVESTIGATION

Forensic Services Division

Microanalysis Standard Operating Procedures Manual

Gunshot Primer Residue Analysis

5. Appropriate measures will be taken to avoid contamination in the laboratory. Personal Protective Equipment (PPE) shall be worn when processing GSR evidence. Prior to processing exhibits, fresh gloves and a clean lab coat shall be worn. The work surface will be cleaned with a detergent solution followed by a 2% hydrochloric acid solution and then clean, unused paper placed on this surface. This same cleaning shall be performed after processing an exhibit. Note: After processing an exhibit with a biohazard, the surface shall be cleaned with a bleach solution first.

4.5. Limitations

Antimony, barium, and lead must be identified using the elemental profile recalled in the EDS software. There are possible peak overlaps of:

Antimony (Sb) with Calcium (Ca), Potassium (K)
Barium (Ba) with Titanium (Ti)
Lead (Pb) with Sulfur (S) and Molybdenum (Mo).

These do not preclude identification, but must be taken into consideration when identifying specific peaks/patterns with specific elements.

4.6. Procedure

When both a hand kit and clothing from a subject are submitted to the laboratory for GSR analysis, the hand kit will be analyzed first. If the results of the analysis are positive, the clothing from the same subject will not be analyzed. However, if the results of the analysis of the hand kit are inconclusive, consistent, or negative, the clothing will be analyzed.

Hand Kits

A new sample stub with carbon tape or adhesive tab (negative control) shall be included with each batch to be analyzed. This stub is to be exposed to the same processes which the evidence sample stubs experience.

Hand kits may be processed as single kits or as batches of kits. Kits from different individuals may be processed in the same batch.

TENNESSEE BUREAU OF INVESTIGATION

Forensic Services Division



Microanalysis Standard Operating Procedures Manual

Gunshot Primer Residue Analysis

Clean the work surface with a detergent solution followed by a 2% hydrochloric acid solution. Place clean, unused paper on the surface.

Put on a clean lab coat and fresh gloves.

Open the kit and document the contents on the GSR Analysis Worksheet. Place a copy of any paperwork present in the kit contents in case file.

Once all kits in the batch have been opened and documented, transfer stub vials to clean area at the carbon coater.

Open one vial at a time. Mark the bottom of the stub with a sample identifier. Place the stub in the coater chamber. Repeat for each vial in the kit. Change gloves after each kit. Repeat for each kit in the batch.

Items/Clothing

A new sample stub with carbon tape or adhesive tab (negative control) shall be included with each batch to be analyzed. This stub is to be exposed to the same processes which the evidence sample stubs experience.

Exhibit items that have been recovered from different locations or persons shall be processed and analyzed separately.

Exhibit items that have been recovered from the same location or the same person may be processed successively without surface cleaning or changing lab coats, and may be analyzed together. Exhibits that contain multiple items in one container may be processed as one item.

Clean the work surface with a detergent solution followed by a 2% hydrochloric acid solution. Place clean, unused paper on the surface.

Put on a clean lab coat and fresh gloves.

Remove the evidence from its original container. Evidence may be photographed for documentation. Large evidence items, such as



TENNESSEE BUREAU OF INVESTIGATION

Forensic Services Division

Microanalysis Standard Operating Procedures Manual

Gunshot Primer Residue Analysis

doors, may be processed differently due to their size. Document changes in case notes.

Prepare new sample stubs with carbon tape or adhesive tabs.

Press the sample stub against the evidence item. This is repeated across the area of collection until the tape/tab is no longer sticky. If the tape/tab is no longer sticky before the collection area is completed, multiple stubs may be used to completely collect the area.

Mark case and stub identifiers on the underside of the stub(s).

Place stub in storage container for transfer to coater.

Return evidence to its original container.

Remove lab coat and gloves.

Clean the work surface with a detergent solution followed by a 2% hydrochloric acid solution. Place clean, unused paper on the surface.

Process the next exhibit as above.

Transfer sample stubs in storage containers to clean area of carbon coater and place the stubs in the coater chamber.

All Samples

Once all sample stubs are placed into the carbon coater chamber, place the chamber under vacuum and coat with a thin film of carbon to increase electrical conductivity. This process is outlined in the Denton carbon coater instructions and is also available in step-by step instructions next to the coater. Return the chamber to atmospheric pressure and remove the stubs.

Bring the SEM/EDS chamber to atmospheric pressure and load sample stubs into the chamber. Document the location of stubs in sample holder. Return the chamber to high vacuum.

Optimize the SEM/EDS. This optimization is to be run with each batch of sample stubs. Optimization will be performed using the

TENNESSEE BUREAU OF INVESTIGATION
Forensic Services Division



Microanalysis Standard Operating Procedures Manual
Gunshot Primer Residue Analysis

parameters below. Cobalt will be the calibration element and is retained on a standard reference stub in the SEM/EDS chamber. The reference material is the MAC Reference Standard for X-ray Microanalysis #7019. Using the GSR software, open a project to store data. On the SEM, once the calibration element is displayed and in proper focus, the acquisition can begin with the EDS software. The SEM spot size should be set to approximately 25% dead time. The spot size can be changed by selecting the 'gun' folder. The acquisition will then be stopped and restarted and allowed to acquire for the full time. The optimization must be run twice. Using the EDS software, select 'measure' and note the value displayed. The value should be 100% +/- 3%. If not within the 3%, repeat the optimization. Generate a spectrum and record the measured value on the spectrum. These results are stored in each case file. The instrument parameters are printed and shall be placed in each case file. These are the operating parameters for the Zeiss(EVO) SEM/EDS:

High Voltage: 20 KeV or 25 KeV
 Beam Current: 30 or 100 uA
 Working Distance: 8.5 mm
 Image Detector: Back Scatter Electron
 Magnification: may vary from 300-500
 Smallest feature: variable
 Method used: GSR with Filters

Method Summary:

SbBaPb	Sb	Ba	Pb	Ca
and/or	0.0-100% 5%-100%	0.0%-100% 5%-100%	0.0%-100% 5%-100%	-----
SbBa	0.0%-100%	0.0%-100%	-----	-----
SbPb	0.0%-100%	-----	0.0%-100%	-----
BaPb	-----	0.0%-100%	0.0%-100%	-----
BaCaPb	-----	0.0%-100%	0.0%-100%	0.0%-100%

The positive control or gunshot primer residue calibration standards (Plano standard, European Network of Forensic Scientist Institute (ENFSI) standard, or sample prepared from known GSR and

TENNESSEE BUREAU OF INVESTIGATION

Forensic Services Division



Microanalysis Standard Operating Procedures Manual

Gunshot Primer Residue Analysis

subjected to repeated analysis to establish the distribution and compositions of the particles) must be first analyzed to prepare the SEM/EDS for the search of high atomic mass number particles. This same sample must be run at the end of each batch.

The negative control stub prepared for each batch shall be run following the pre-analysis positive control and prior to the post-analysis positive control for that batch. If all sample stubs in the batch are from the same individual or location then the negative control stub may be run only at the beginning of the batch.

If the analysis of the positive control sample does not detect one (1) micrometer particles or larger, the results from the batch of sample stubs will not be used. Optimize the SEM/EDS hardware and software to detect one (1) micrometer particles and larger. Reanalyze the batch of sample stubs.

For stubs that indicate the presence of particles of gunshot primer residue, the analyst will return to these particles on the stub and manually re-acquire spectra and verify proper morphology to confirm their identity. It is not necessary to confirm every particle; however, a number of particles must be confirmed. The hard copy printout becomes part of the case notes.

When analysis is completed, the stubs may then be removed from the sample chamber of the SEM and placed into their designated containers which have been labeled with the laboratory case number, examiner's initials and exhibit number. This container can be placed in with the original evidence or be returned separately.

The negative control sample(s) for items/clothing will be returned with the evidence in its own separate container. Negative control stubs on batch analysis cannot be returned and will not be retained.

4.7. Interpretation

Particles that are *identified* as Gunshot Primer Residue must contain all three elements: antimony, barium and lead. In the total population of suspected particles, multiple (two or more) particles with all elements of GSR must be present. Additionally, particles with two of the three elements of GSR may also be present in the total population. All of these particles shall have morphology consistent with a high temperature

TENNESSEE BUREAU OF INVESTIGATION

Forensic Services Division



Microanalysis Standard Operating Procedures Manual

Gunshot Primer Residue Analysis

vapor condensing into solid droplets. The size of these particles, range from approximately 0.5 micrometers to approximately 100+ micrometers.

Particles considered to be *consistent* with Gunshot Primer Residue must have morphology as described above and have one of the following elemental profiles:

- antimony, barium (with no more than a trace of iron or sulfur)
- lead, barium
- lead, antimony (with levels of antimony greater than trace amounts)

Additionally, multiple (two or more) particles must be present to be considered consistent with Gunshot Primer Residue. This can include one three component particle and one two component particle. Particles that don't meet the above criteria will not be considered Gunshot Primer Residue.

Instruments and Equipment

1. Vacuum Carbon Coater, carbon and supplies
2. Scanning Electron Microscope with Energy Dispersive Spectrometer
3. SEM aluminum sampling stubs
4. Double-sided conductive tape or "sticky" tabs
5. Photographic equipment with accessories
6. SEM stub evidence containers

Measurement Traceability

The INCA software is optimized to detect 1 micrometer or larger GSR particles. This measurement is a detection threshold only. The size of the particle is irrelevant to the determination of GSR. Determination is based on presence of components and morphology of the particle.

Reference Materials

1. Nickel Grid
2. Plano Standard
3. Cobalt Standard
4. ENFSI/Plano/Known GSR standard

Reports

The following are possible results concluded from the examination:

TENNESSEE BUREAU OF INVESTIGATION

Forensic Services Division



Microanalysis Standard Operating Procedures Manual

Gunshot Primer Residue Analysis

For Items:

Electron microscopic examination and analysis of exhibit ____ revealed the presence of particles identified as gunshot primer residue. Individual microscopic particles of gunshot primer residue have the appearance of being condensed from a vapor and contain the elements of antimony, barium and lead. These results indicate that this exhibit was near a gun when it was fired, came in contact with a recently fired gun and/or recently fired ammunition components.

Electron microscopic examination and analysis of exhibit ____ revealed the presence of a single particle identified as gunshot residue. Individual microscopic particles of gunshot residue have the appearance of being condensed from a vapor and contain the elements antimony, barium, and lead. A single particle of gunshot residue is insufficient to determine if this exhibit was near a gun when it was fired, came in contact with a recently fired gun and/or recently fired ammunition components.

Electron microscopic examination and analysis of this exhibit revealed the presence of particles consistent with gunshot primer residue. These consistent particles are individual microscopic particles and have the appearance of being condensed from a vapor. However, they contain only two of the three elements (antimony, barium and lead) required to identify them as gunshot primer residue. These particles are found in gunshot primer residue, but may originate from other occupational and/or industrial sources as well. These results cannot eliminate the possibility that this exhibit was near a gun when it was fired, came in contact with a recently fired gun, and/or came in contact with fired ammunition components.

Electron microscopic examination and analysis of exhibit ____ did not reveal the presence of particles of gunshot primer residue. The absence of gunshot primer residue is consistent with an item not being exposed to a source of gunshot primer residue. A negative result could also occur when gunshot primer residue particles are lost due to washing, excessive time interval between firearm discharge and collection, and/or other routine activities.

Hands:

Electron microscopic examination and analysis of exhibit ____ revealed the presence of particles identified as gunshot primer residue. Individual microscopic particles of gunshot primer residue have the appearance of being condensed from a vapor and contain the elements of antimony, barium and lead. These results indicate that the individual could have fired, handled or was near a gun when it fired.



TENNESSEE BUREAU OF INVESTIGATION

Forensic Services Division

Microanalysis Standard Operating Procedures Manual

Gunshot Primer Residue Analysis

Electron microscopic examination and analysis of exhibit ____ revealed the presence of a single particle identified as gunshot residue. Individual microscopic particles of gunshot residue have the appearance of being condensed from a vapor and contain the elements antimony, barium, and lead. A single particle of gunshot residue is insufficient to determine if the individual could have fired, handled, or was near a gun when it fired.

Electron microscopic examination and analysis of exhibit ____ revealed the presence of particles consistent with gunshot primer residue. These consistent particles are individual microscopic particles and have the appearance of being condensed from a vapor. However, they contain only two of the three elements (antimony, barium and lead) required to identify them as gunshot primer residue. These particles are found in gunshot primer residue, but may originate from other occupational and/or industrial sources as well. These results cannot eliminate the possibility that the individual could have fired, handled or was near a gun when it fired.

Electron microscopic examination and analysis of exhibit ____ did not reveal the presence of particles of gunshot primer residue. The absence of gunshot primer residue is consistent with an individual not having fired a weapon or being exposed to a source of gunshot primer residue. A negative result could also occur when gunshot primer residue particles are lost due to washing, excessive time interval between firearm discharge and collection, and/or other routine activities.

Electron microscopic examination and analysis of exhibit ____ did not reveal the presence of particles of gunshot primer residue. It must be noted that some .22 ammunition does not have all the elements needed for gunshot residue identification. These results cannot eliminate the possibility that the individual could have fired, handled, or was near a gun when it fired.

However, it should be noted that the time between firearm discharge and collection was excessive.

The wording of these results may vary.