



Designation: ~~E2458 – 10~~ E2458 – 17

Standard Practices for Bulk Sample Collection and Swab Sample Collection of Visible Powders Suspected of Being BiothreatBiological Agents and Toxins from Nonporous Surfaces¹

This standard is issued under the fixed designation E2458; the number immediately following the designation indicates the year of original adoption or, in the case of revision, the year of last revision. A number in parentheses indicates the year of last reapproval. A superscript epsilon (ϵ) indicates an editorial change since the last revision or reapproval.

1. Scope

1.1 These practices address collection of visible powders that are suspected biothreatbiological agents and toxins from solid nonporous surfaces using a bulk collection method, using a dry swab and laminated card, followed by a swab sampling method using a sterile moistened swab. Bulk powder samples are collected and packaged in a manner that permits the maximum amount of the sample to be safely transported to a reference laboratory within the Centers for Disease Control and Prevention (CDC) national Laboratory Response Network (LRN)² for confirmatory identification and safe storage. If the source of the powder is a letter or small package, that item is also packaged in a manner that permits it to be safely transported to an LRN reference laboratory. A sterile moistened swab may be used to collect residual powder from the nonporous surface and may be used to conduct on-site biological assessments for the purpose of testing for biothreatagentsbiological agents and toxins.

1.2 These practices are performed in coordination with the Federal Bureau of Investigation (FBI) as part of a risk assessment including hazard assessment and threat credibility evaluation as recommended and clarified in Guide E2770. The decision to implement these practices and collect a public safety sample will be made by members of the response community of the jurisdiction assuming responsibility through coordination with the FBI and the receiving LRN reference laboratory.

1.3 Sample Collection Method A covers the bulk collection and packaging of suspicious visible powders that are suspected biothreatbiological agents and toxins from solid nonporous surfaces. All samples suspected to be biothreatbiological agents and toxins on nonporous surfaces should be collected according to Sample Collection Method A and sent to an LRN reference laboratory for confirmatory testing.

1.4 Sample Collection Method B covers swab sampling of residual suspicious powders that are suspected biothreatbiological agents and toxins from solid nonporous surfaces. Swab samples can be used for on-site biological assessment; however results from on-site biological assessments are not definitive; confirmatory testing by the LRN reference laboratory is necessary to make public health decisions.

1.5 These practices incorporate reference guidance for packaging and transport of suspicious visible powders to comply with all appropriate federal regulations regarding biosafety and biosecurity.

1.6 These practices should only be used to collect visible samples that are suspected biothreatbiological agents and toxins and have been field screened according to reference guidance for explosive hazard, radiological hazard, and other acute chemical hazards.

1.7 The values stated in SI units are to be regarded as standard. The values given in parentheses are for information only.

1.8 *This standard does not purport to address all of the safety concerns associated with its use. It is the responsibility of the user of this standard to establish appropriate safety and health practices and determine the applicability of regulatory limitations prior to use.*

1.9 *This international standard was developed in accordance with internationally recognized principles on standardization established in the Decision on Principles for the Development of International Standards, Guides and Recommendations issued by the World Trade Organization Technical Barriers to Trade (TBT) Committee.*

¹ These practices are under the jurisdiction of ASTM Committee E54 on Homeland Security Applications and are the direct responsibility of Subcommittee E54.01 on CBRNE Sensors and Detectors.

Current edition approved Oct. 1, 2010 May 15, 2017. Published November 2010 June 2017. Originally approved in 2006. Last previous edition approved in 2006 2010 as ~~E2458 – 06~~ E2458 – 10. DOI: ~~10.1520/E2458-10~~ 10.1520/E2458-17.

² The CDC Laboratory Response Network is the network responsible for handling clinical specimens and environmental samples containing suspected biothreat agents.

2. Referenced Documents

2.1 ASTM Standards:³

E2770 Guide for Operational Guidelines for Initial Response to Suspected Biological Agents and Toxins
 F2412 Test Methods for Foot Protection
 F2413 Specification for Performance Requirements for Protective (Safety) Toe Cap Footwear

2.2 Federal Government Regulations:⁴

18 USC ~~175~~178 Prohibitions with respect to biological weapons
 DOT 49 CFR, Parts 171-180 Hazardous Materials Regulations
 DOT - 49 CFR 172 Subpart H, Transportation Training
 DOT - 49 CFR 173 General Requirements for Shipments and Packagings
 DOT - 49 CFR 178 Specifications for Packagings
 EPA - 40 CFR 300 National Oil and Hazardous Substances Pollution Contingency Plan (NCP)
 EPA - 40 CFR 311 Worker Protection
 NRC - 10 CFR 20 Standards for Protection against Radiation
 NIOSH - 42 CFR 84 Respiratory Protective Devices
 OSHA - 29 CFR 1910 Subpart Z and 29 CFR 1926 Subpart Z Toxic and Hazardous Substances
 OSHA - 29 1910.1096 and 29 CFR 1926.53 Ionizing Radiation
 OSHA - 29 CFR 1910.120 Hazardous Waste Operations and Emergency Response (HAZWOPER) standard
 OSHA - 29 CFR 1910 Subpart I (Sections 132 to 139) Personal Protective Equipment
 OSHA - 29 CFR 1910.1200 Hazard Communication

2.3 Federal Guidance:

OSHA - CPL 02-02-073 Inspection Procedures for 29 CFR 1910.120 and 1926.65, Paragraph (q): Emergency Response to Hazardous Substance Releases
 NIOSH Publication No. 2009-132 Recommendations for the Selection and Use of Respirators and Protective Clothing for Protection Against Biological Agents
 FBI Laboratory Publication Handbook of Forensic Services 20032013
 FBI-DHS-HHS/CDC Coordinated Document Guidance on Initial Response to a Suspicious Letter/Container with a Potential Biological threat, November 2, 2004
 CDC/NIOSH Surface Sampling Procedures for *Bacillus anthracis* Spores from Smooth, Non-porous Surfaces, April 26, 2012⁵
 DHS Framework for a Biothreat Field Response Mission Capability, April 2011⁶
 Sandia National Laboratories SAND2005-3237 (LBNL-54973 (II)) Guidelines to Improve Airport Preparedness Against Chemical and Biological Terrorism⁷

2.4 NFPA Standards:⁸

NFPA 472 Standard for Competence of Responders to Hazardous Materials/Weapons of Mass Destruction Incidents, 20082013 Edition
 NFPA 1994 Standard on Protective Ensembles for Chemical/Biological Terrorism Incidents

2.5 IATA Standards:⁹

IATA PI 602 Infectious Diseases (Infectious Substances)
 IATA PI 650 Shipping of Diagnostic Samples
 IATA DGR 46th Edition, 2005
 IATA DGR Addendum I, January 2005
 IATA DGR Addendum II, March 2005
 IATA DGR Addendum III, July 2005

2.6 ANSI Standards:¹⁰

ANSI Z41-1999 American National Standard for Personal Protection - Protective Footwear
 ANSI Z87.1-2003 American National Standard for Occupational and Educational Personal Eye and Face Protection Devices
 ANSI Z88.2-1992 American National Standard Practices for Respiratory Protection
 ANSI Z88.10-2001 American National Standard for Personal Protection - Respirator Fit Testing Methods

³ For referenced ASTM standards, visit the ASTM website, www.astm.org, or contact ASTM Customer Service at service@astm.org. For *Annual Book of ASTM Standards* volume information, refer to the standard's Document Summary page on the ASTM website.

⁴ Available from U.S. Government Printing Office Superintendent of Documents, 732 N. Capitol St., NW, Mail Stop: SDE, Washington, DC 20401, and also available online from Occupational Safety and Health Administration (www.osha.gov).

⁵ Available from <http://www.cdc.gov/niosh/topics/emres/surface-sampling-bacillus-anthraxis.html>.

⁶ Available from <http://www.hsdl.org/?view&did=767721>.

⁷ Available from <http://share-ng.sandia.gov/news/resources/releases/2005/images/unlsand-2005-3237.pdf>.

⁸ Available from National Fire Protection Association (NFPA), 1 Batterymarch Park, Quincy, MA 02269-9101.

⁹ Available from the International Air Transport Association, 800 Place Victoria, PO Box 113, Montreal-H4Z 1M1, Quebec, Canada.

¹⁰ Available from American National Standards Institute (ANSI), 25 W. 43rd St., 4th Floor, New York, NY 10036, <http://www.ansi.org>.

ANSI/ISEA Z89.1-2003 American National Standard for Personal Protection - Protective Headwear for Industrial Workers Requirements

ANSI/Compressed Gas Association, CGA G-7.1-1997 Commodity Specification for Air

2.7 IAFC Guidance:¹¹

Model Procedures for Responding to a Package with Suspicion of a Biological Threat, October 2008

3. Terminology

3.1 Definitions:

3.1.1 *aseptic technique*, *n*—operation or performance of a procedure or method under carefully controlled conditions to reduce the risk of exposure and prevent the introduction of unwanted material/matter (contamination) into a sample.

3.1.2 ~~*biothreatbiological agent*, *n*—any microorganism, virus, infectious substance, or biological product that may be engineered as a result of biotechnology, or any naturally occurring or bioengineered microorganism (including but not limited to, bacteria, viruses, fungi, rickettsiae, or protozoa), or infectious substance, or any naturally occurring, bioengineered or synthesized component of any such microorganism, virus, microorganism or infectious substance, or biological product, capable of causing: (1) death, disease or other biological malfunction in a human, an animal, a plant, or another living organism; (2) deterioration of food, water, equipment, supplies, or material of any kind; or (3) deleterious alteration of the environment.~~ **18 USC 175178**

3.1.3 *bulk powder*, *n*—a visible powder, at least approximately ~~1 teaspoon or 5 ml~~ 5 mL (1 teaspoon) in volume amassed or dispersed over a limited area (optimally, area should be less than 20 by 20 cm (approximately 8 by 8 in.)).

3.1.4 *chain of custody*, *n*—set of procedures and documents to account for the integrity of sample by tracking its handling and storage from point of sample collection to final disposition of the sample.

3.1.5 *cold zone*, *n*—the uncontaminated area where workers are unlikely to be exposed to hazardous substances or dangerous conditions; also known as Clean Zone or Support Zone. **CPL 02-02-071 Directive**

3.1.6 *confirmatory analysis*, *n*—a test or a series of assays that definitively identifies the presence of a suspected substance or agent.

¹¹ Available from International Association of Fire Chiefs (IAFC), 4025 Fair Ridge Drive, Suite 300 Fairfax, VA 22033, <http://www.iafc.org>.

3.1.6.1 Discussion—

Confirmatory analysis of a ~~biothreat~~ biological agent for public health action can be performed only by an LRN national or reference laboratory.

3.1.7 *decontamination*, *n*—the physical or chemical process, or both, of reducing and preventing the spread of contaminants from people, animals, the environment, or equipment involved at hazardous materials/weapons of mass destruction (WMD) incidents. **NFPA**

3.1.8 *field screening*, *n*—field measurements utilized early in the ~~site assessment process~~ response to define and ~~delineate~~ characterize the ~~contaminants present, potential hazards present, including corrosive, flammable, volatile, radioactive, or oxidizer hazards, and to support tactical decision making and to~~ address operational safety measures.

3.1.8.1 Discussion—

Field screening does not include measurements of biological properties, which is termed on-site biological assessments (see 3.1.12).

3.1.9 *hazard*, *n*—something that is potentially dangerous or harmful, often the root cause of an unwanted outcome; a danger or peril. **NIMS**

3.1.10 *hot zone*, *n*—the area, located on the site where contamination is either known or expected and where potential for greatest exposure exists; also known as Exclusion Zone or ExZ. **CPL 02-02-071 Directive**

3.1.11 *incident commander (IC)*, *n*—the individual responsible for all incident activities, including the development of strategies and tactics and the ordering and release of resources.

3.1.11.1 Discussion—

The IC has overall authority and responsibility for conducting incident operations and is responsible for the management of all incident operations at the incident site. **NIMS**

3.1.12 *on-site biological assessment*, *n*—measurements of properties inherent to biological materials performed in the field using rapid, field-based procedures and assays.

3.1.13 *personal protective equipment (PPE)*, *n*—equipment provided to shield or isolate a person from the chemical, biological, physical, and thermal hazards that can be encountered at hazardous materials/weapons of mass destruction (WMD) incidents. **NFPA**

3.1.14 *presumptive test*, *n*—non-definitive test used to evaluate a material for the presence of a substance or agent, or the presence of signatures of a substance or agent.

3.1.15 *risk*, *n*—the probability of suffering a loss or harm or injury; peril.

3.1.16 *toxin*, *n*—the toxic material or product of plants, animals, microorganisms (including but not limited to, bacteria, viruses, fungi, rickettsiae, or protozoa), or infectious substances, or a recombinant or synthesized molecule, whatever their origin and method of production, and includes: (1) any poisonous substance or biological product that may be engineered as a result of biotechnology produced by a living organism; or (2) any poisonous isomer or biological product, homolog, or derivative of such a substance (18 USC 178).

3.1.17 *threat*, *n*—an indication of possible violence, harm, or danger and may include an indication of intent and capability. **NIMS**

3.1.18 *warm zone*, *n*—the transition area between the Exclusion Zone (ExZ or hot zone) and the Support Zone (SZ or cold zone) used to reduce and limit the amount of contamination on people and equipment, and in the air, water, and soil that may be transferred into nonhazardous areas; the CRZ contains decontamination facilities, and functions as a buffer zone surrounding the ExZ; also known as the contamination reduction zone or CRZ. **CPL 02-02-071 Directive**

3.1.19 *weapon of mass destruction (WMD)*, *n*—any weapon or device that is intended, or has the capability, to cause death or serious bodily injury to a significant number of people through the release, dissemination, or impact of (1) toxic or poisonous chemicals or their precursors; (2) a disease organism; or (3) radiation or radioactivity. **U.S. Code Title 50, Ch. 40, Sect. 2302, War and National Defense Definitions**

3.2 Acronyms:

- 3.2.1 *AHJ*—Authority Having Jurisdiction
- 3.2.2 *ANSI*—American National Standards Institute
- 3.2.3 *ASTM*—American Society for Testing and Materials
- 3.2.4 *CDC*—Centers for Disease Control and Prevention
- 3.2.5 *CFR*—Code of Federal Regulations
- 3.2.6 *CRZ*—Contamination Reduction Zone
- 3.2.7 *CST*—Civil Support Team
- 3.2.8 *DHS*—Department of Homeland Security
- 3.2.9 *DOT*—Department of Transportation
- 3.2.10 *EOC*—Emergency Operations Center
- 3.2.11 *EPA*—Environmental Protection Agency
- 3.2.12 *ExZ*—Exclusion Zone
- 3.2.13 *FBI*—Federal Bureau of Investigation
- 3.2.14 *FEMA*—Federal Emergency Management Agency
- 3.2.15 *HAZMAT*—Hazardous Materials
- 3.2.16 *HHS*—Health and Human Services
- 3.2.17 *IAFC*—International Association of Fire Chiefs
- 3.2.18 *IATA*—International Air Transport Association
- 3.2.19 *IC*—Incident Commander
- 3.2.20 *ICS*—Incident Command System
- 3.2.21 *IEC*—International Electrotechnical Commission
- 3.2.22 *ISEA*—International Safety Equipment Association
- 3.2.23 *ISO*—International Organization for Standardization
- 3.2.24 *LRN*—Laboratory Response Network
- 3.2.25 *MACS*—Multiagency Coordination System
- 3.2.26 *NFPA*—National Fire Protection Association

- 3.2.27 *NIMS*—National Incident Management System
- 3.2.28 *NIOSH*—National Institute for Occupational Safety and Health
- 3.2.29 *NRC*—Nuclear Regulatory Commission
- 3.2.30 *OSHA*—Occupational Safety and Health Administration
- 3.2.31 *PBS*—Phosphate Buffered Saline
- 3.2.32 *PPE*—Personal Protective Equipment
- 3.2.33 *SZ*—Support Zone
- 3.2.34 *USAR*—*US&R*—Urban Search and Rescue
- 3.2.35 *WMD*—Weapons of Mass Destruction

4. Significance and Use

4.1 These practices should be used only to collect visible samples that are suspected ~~biothreat~~ biological agents and toxins and have been field screened as defined by the FBI-DHS-HHS/CDC Coordinated Document for explosive hazard, radiological hazard, and other acute chemical hazards.

4.2 These practices provide standardized methods for collecting, packaging, and transporting suspicious visible powder samples that are suspected ~~biothreat agents~~ biological agents and toxins. Collection of a bulk powder material from a nonporous surface using a sterile swab and laminated card as the collection devices to move the material into a container will depend on several factors, including (but not limited to): (1) amount of visible powder present; (2) sample composition; (3) choice of collection device; (4) size and shape of the collection container; (5) ability of the powder to become aerosolized; (6) texture and porosity of the surface; (7) humidity; (8) air movement; and (9) electrostatic properties of powders and collection tools/containers.

4.3 Similarly, these practices standardize methods for sampling suspicious visible powders for on-site analysis, although wipe and swab sampling is often employed in the field for subsequent LRN reference laboratory analysis. The ability to collect suitable samples from nonporous surfaces using a sterile moistened swab will depend on the following factors: (1) swabbing procedure; (2) swab material; (3) sample composition; and (4) texture of the surface.

4.4 These practices standardize suspicious powder collection and packaging procedures and swab sampling procedures in order to reduce exposure risk, to reduce variability associated with sample handling and sample analysis, and to increase reliability of sampling visible powder samples from nonporous surfaces.

4.5 The bulk sample collection practice and the swab sampling practice are recommended for collecting amassed or dispersed powder samples from all nonporous surfaces on which the suspicious powder sample is clearly visible.

4.6 These practices are not recommended for samples on porous materials such as upholstery, carpeting, air filters, or ceiling tiles.

4.7 These practices are recommended for collecting visible powders where the bulk of the powder sample is amassed or dispersed over a limited area (optimally, area should be less than 20 by 20 cm (approximately 8 by 8 in.) or 400 cm² (approximately 64 in.²).

4.8 These practices are to be performed by personnel who are adequately trained to work with hazardous materials in the hot zone (see NFPA 472, or OSHA - 29 CFR 1910.120). Personnel performing collection or screening under these practices shall be adequately trained in the use of sampling equipment, materials, and procedures. This includes personnel performing the prior initial chemical and radiological screening. Personnel should use the appropriate level of personal protective equipment (PPE) to mitigate hazards during collection and screening. Personnel performing collection or screening under these practices shall be aware of evidence preservation and sampling procedures (NFPA 472 section 6.5).

4.9 These standard practices should be used in accordance with Guide [E2770](#) for best practices for planning, training and evaluation of competency.

SAMPLE COLLECTION METHOD A—BULK SAMPLE COLLECTION FOR LABORATORY ANALYSIS

5. Scope of Method A

5.1 This sample collection method applies to the bulk collection and packaging of suspicious visible powders that are suspected ~~biothreat agents~~ biological agents or toxins, or both, from solid nonporous surfaces.

5.2 These practices are performed in coordination with the FBI and receiving LRN reference laboratory after a risk assessment including a hazard assessment and threat credibility evaluation is conducted and the sample is deemed potentially to be a credible threat as recommended and clarified in Guide [E2770](#).

5.3 This sample collection method applies to suspicious visible powders that are amassed or dispersed in a limited area where the bulk of the powder sample is in an area that is less than 20 by 20 cm (approximately 8 by 8 in.) or 400 cm² (approximately 64 in.²).

5.4 These practices should be used only to collect samples that are suspected ~~biothreat hazards and biological agent and toxin hazards~~ that have been field screened as defined by the FBI-DHS-HHS/CDC Coordinated Document for explosive, radiological, and chemical hazards.

6. Summary of Sample Collection Method A

6.1 A visible powder sample that is a suspected ~~biothreat agent~~ biological agent or toxin, or both, and its source should be field screened for non-biological hazards as defined in the FBI-DHS-HHS/CDC Coordinated Document and according to the reference guidance including appropriate NFPA documents. Non-biological hazards include explosive, radiological, and chemical hazards. The visible powder sample, amassed or dispersed in a limited area, is collected from a nonporous surface using a swab and a laminated card to move the sample into a sterile dry collection container that is held close to the surface.

6.2 The method provides guidance on performing these procedures in a manner that will minimize sample loss and aerosolization of the powder. The bulk powder sample and swab are sent to ~~an~~ LRN reference laboratory for confirmatory analysis.

7. Sampling and Packaging Equipment and Supplies

7.1 *Personal Protective Equipment*—Level A, B, or C personal protective equipment ensembles as indicated (see Section 2 for additional guidance, including OSHA - 29 CFR 1910.120 Appendix B and NFPA 1994).

7.2 Clean drop cloth to create a ~~clean~~ isolated work area.

7.3 *Sample Transport Container*—Bucket or large heavy duty plastic ~~bag~~ used to transport the samples and materials from the hot zone through to the decontamination line.

7.4 *Non-powdered Nitrile or Vinyl Examination Gloves.*

7.5 *Two Sterile Polypropylene Wide-mouth Screw-capped Sample Collection Containers (Sample Containers)*—Containers must possess a leak-resistant seal; Diameter of container mouth must be large enough to accommodate the 4 by 6.5 cm plastic cards (Section 7.10); each pre-labeled as “POWDER SAMPLE” with unique sample identifier numbers.

7.6 *Bleach Solution*—Fresh pH-adjusted bleach solution (household bleach diluted 1:9; pH-adjusted to 6.8-8.0) to be prepared outside of the hot zone just prior to use by (step 1) mixing one part household bleach (5.25 to 6.0 % sodium hypochlorite) with 5 parts water ~~(+/-);(volume/volume)~~; (step 2) adding 1 part white vinegar; (step 3) adding 3 parts of additional water.¹²

7.7 *Labeling or Marking Tape.*

7.8 *Solvent-resistant Indelible Marker.*

7.9 *Sterile Culture Swabs*, (rayon, macrofoam, or polyester) individually packaged and sterile, self-contained in sealed plastic tubes, with absorbent material wrapped around one end of a plastic stick, unopened.

7.10 *Two at least 4 by 6.5 cm (approximately 1.5 by 2.5 in.) Sterile Plastic Laminated Cards.*

7.11 *Six 1-gal Sealable Plastic Bag(s)*—Both bags with sliding lock and Trademark Whirl-Pak¹³ bags are not recommended for this purpose since they may not seal completely; the use of colored or opaque bags is discouraged because it makes viewing of the sample more difficult once transported back to the laboratory in the CDC Laboratory Response Network.

7.12 *Bucket*, or other container marked “BIOHAZARD WASTE”.

7.13 *1-gal Sealable Plastic Bag(s)*, pre-labeled as “BIOHAZARD WASTE”.

7.14 *Field Screening Results Form*—See example in **Appendix X1**.

7.15 *Sample Collection Sheet*—See example in **Appendix X1**. Note that a single sample collection sheet can be used for all items collected at a single location including primary source, swab, and powder sample(s).

7.16 *Chain-of-Custody Form*—See example in **Appendix X1**.

7.17 *Two Plastic Transparent Document Pouches*, with adhesive on back.

7.18 *Durable Hard-sided Outer Container*, with lid or screw cap, for sample transport (such as metal can with lid, or plastic container with lid).

¹² Additional information about decontamination with pH-adjusted bleach solution is available at www.epa.gov/opp00001/factsheets/chemicals/bleachfactsheet.htm.
nepis.epa.gov.

¹³ Whirl-Pak is a trademark of Nasco International, Inc.