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AnAmerican National Standard

Standard Practice for Personal Sampling and Analysis of Endotoxin in Metalworking Fluid Aerosols in Workplace Atmospheres[1]

This standard is issued under the fixed designation E2144; 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 This practice covers quantitative methods for the personal sampling and determination of bacterial endotoxin concentrations in poly-disperse metalworking fluid aerosols in workplace atmospheres. Users should have fundamental knowledge of microbiological techniques and endotoxin testing.

1.2 Users of this practice may obtain personal or area exposure data of endotoxin in metalworking fluid aerosols, either on a short-term or full-shift basis in workplace atmospheres.

1.3 This practice gives an estimate of the endotoxin concentration of the sampled atmosphere.

1.4 This practice seeks to minimize inter laboratory variation but does not ensure uniformity of results.

1.5 It is anticipated that this practice will facilitate inter laboratory comparisons of airborne endotoxin data from metalworking fluid atmospheres, particularly metal removal fluid atmospheres, by providing a basis for endotoxin sampling, extraction, and analytical methods.

1.6 In 1997, the Occupational Safety and Health Administration (OSHA) empanelled a Standards Advisory Committee to make recommendations to the Administration regarding measures that the Administration could take to improve the health of workers exposed to metalworking fluids. A report to the Assistant Secretary of Labor for OSHA was submitted in July, 1999. Subcommittee E34.50 believes that the user community would benefit significantly if a standard method was developed to give the community guidance on a methodology for the sampling and analysis of personal airborne endotoxin exposure assessments in facilities using water-miscible metal removal fluids, based on the LAL assay or other endotoxin detection technologies as they become available.

1.7 This practice does not attempt to set or imply limits for personal exposure to endotoxin in metalworking fluid aerosols in workplace environments.

1.8 *This standard does not purport to address all of the safety concerns, if any, 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.*

2. Referenced Documents

2.1 *ASTM Standards:*[2]

D1356 Terminology Relating to Sampling and Analysis of Atmospheres

D4840 Guide for Sample Chain-of-Custody Procedures

D5337 Practice for Flow Rate Adjustment of Personal Sampling Pumps

D6629 Guide for Selection of Methods for Estimating Soil Loss by Erosion

E1370 Guide for Air Sampling Strategies for Worker and Workplace Protection

E1497 Practice for Selection and Safe Use of Water-Miscible and Straight Oil Metal Removal Fluids

E1542 Terminology Relating to Occupational Health and Safety

2.2 *OSHA Standards:*[3]

29 CFR 1910.1000 Air Contaminants

29 CFR 1910.1450 Occupational Exposure to Hazardous Chemicals in Laboratories

2.3 *Other Documents:*

Criteria Document for a Recommended Standard: Occupational Exposure to Metalworking Fluids[4]

NIOSH Manual of Analytical Methods (NMAM)[4]

[1] This practice is under the jurisdiction of ASTM Committee E34 on Occupational Health and Safety and is the direct responsibility of Subcommittee E34.50 on Health and Safety Standards for Metal Working Fluids.

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[2] 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.

[3] Available from U.S. Government Printing Office Superintendent of Documents, 732 N. Capitol St., NW, Mail Stop: SDE, Washington, DC 20401, http://www.access.gpo.gov..

[4] Available from U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, National Institute for Occupational Safety and Health, 4676 Columbia Pkwy., Cincinnati, OH 45226.

3. Terminology

3.1 For definitions of terms in this practice relating to sampling and analysis of atmospheres, refer to Terminology D1356. For definitions of terms in this practice relating to occupational health and safety, refer to Terminology E1542.

3.2 *Definitions of Terms Specific to This Standard:*

3.2.1 *endotoxin, n*—pyrogenic high molar mass lipopolysaccharide (LPS) complex associated with the cell wall of gram-negative bacteria.

3.2.1.1 *Discussion*—Though endotoxins are pyrogens, not all pyrogens are endotoxins. Endotoxins are specifically detected through a Limulus Amoebocyte Lysate (LAL) test.

3.2.2 *endotoxin unit (EU), n*—a biological potency unit equivalent to the FDA Reference Standard Endotoxin (RSE). Currently, EC-6 is equivalent to 0.1 ng 3D 1 EU.

3.2.3 *field blank, n*—filter/cassette unit prepared for sampling that is taken to the sampling site and handled in the same manner as the analytical filter/cassette unit, but that is not a part of the sampling process.

3.2.4 *Gram-negative bacteria, n*—prokaryotic cells that have a complex cell-wall structure that stain characteristically when subjected to the differential Gram staining procedure.

3.2.5 *Limulus amebocyte lysate (LAL) assay, n*—a biological assay that detects endotoxin.

3.2.6 *metal removal fluids, n*—the subset of metal working fluids that are used for wet machining or grinding to produce the finished part.

3.2.6.1 *Discussion*—The term most often refers to straight oils and water-based fluids, such as soluble, semi-synthetic, and synthetic fluids.

3.2.7 *onset time, n*—time required for a change of 200 mOD (optical density) units relative to the initial OD value.

3.2.8 *personal sampler, n*—a portable sampling instrument that is attached to a person to ascertain the concentration of specific constituents in the air in the person's breathing zone.

3.2.9 *pyrogen-free, adj*—material(s) devoid of measurable endotoxin activity.

3.2.10 *pyrogen-free water (PFW), n*—processed water that is devoid of measurable endotoxin activity.

4. Summary of Practice

4.1 A known volume of workplace air in a facility utilizing metalworking fluids is drawn through a sample filter cassette unit.

4.2 The sample filter is extracted into a pyrogen-free solution to quantitatively release endotoxin absorbed from collected metalworking fluid aerosol.

4.3 The extract solution is subjected to quantitative endotoxin analysis techniques. The measured endotoxin concentration is reported in terms of endotoxin potency units per unit volume of air sampled.

5. Significance and Use

5.1 Endotoxins in metalworking fluid aerosols present potential respiratory health hazards to workers who inhale them. Therefore, a consensus standard is needed to provide reliable data on workplace airborne endotoxin concentrations where metalworking fluids are used.

5.2 This practice for measuring airborne endotoxin concentrations in metalworking fluid atmospheres will help to foster a better understanding of endotoxin exposure-response relationships.

5.3 This practice facilitates comparisons of inter laboratory data from methods and field investigative studies.

6. Interferences

6.1 Airborne endotoxin measurements resulting from use of LAL reagents are subject to inhibition/enhancement effects from a variety of bio-molecular species and physicochemical phenomena, such as pH, temperature, filter matrix effects, cationic concentrations, LAL-reactive materials (LRM), enzyme influences, and lysate composition variability and sensitivity (a function of different lysate processing methodologies).

7. Apparatus

7.1 *Sampling:*

7.1.1 *Sampling Unit,* an apparatus consisting of a personal sampling pump, a 37-mm glass fiber filter, a two-piece, closed-face plastic cassette, and flexible connecting tubing between the personal sampling pump and the attached cassette/filter unit.

7.1.1.1 *Pump,* a constant-flow personal sampling pump with an on-board battery power source and a flow rate of 2.0 L/min (±5 %).

7.1.1.2 *Filter Cassette,* pyrogen-free, closed-faced, two-piece polystyrene filter holder with 4 mm inlet and outlet, with caps.

7.1.1.3 *Filter (Membrane),* pyrogen-free, glass fiber, 37-mm diameter with a cellulose support pad.

7.1.1.4 *Connective Tubing,* flexible, appropriate inside diameter.

7.1.1.5 *Soap-bubble Meter,* a primary standard used for sampler flow rate calibration.

NOTE 1—An alternative primary standard is acceptable.

7.2 *Extraction:*

7.2.1 *Sonicator Bath,* ultrasonic/water bath apparatus with a minimum peak frequency of 40-kHz with cavitation adjustment and thermostat control.

7.2.2 *Vortex Mixer,* general purpose with a minimum speed of 500 rpm.

8. Reagents and Materials

8.1 *Control Standard Endotoxin (CSE)*—Endotoxin preparations used for calibration standards shall be referenced to the Federal Drug Administration (FDA) Reference Standard Endotoxin (RSE), which is presently EC-6 RSE. Calibration standards data and corresponding regression data are expressed in EU.

8.2 Endotoxin detection reagents, utilized in accordance with manufacturer's directions.