Journal of ASTM International
Selected Technical Papers

STP 1533

Surface and Dermal Sampling

JAI Guest Editors:

Michael Brisson
Kevin Ashley

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100 Barr Harbor Drive
PO Box C700
West Conshohocken, PA 19428-2959

Printed in the U.S.A.

ASTM Stock #: STP1533

Library of Congress Cataloging-in-Publication Data

ISBN: 978-0-8031-7519-8

Journal of ASTM International (JAI) Scope

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Citation of Papers

When citing papers from this publication, the appropriate citation includes the paper authors, "paper title", J. ASTM Intl., volume and number, Paper doi, ASTM International, West Conshohocken, PA, Paper, year listed in the footnote of the paper. A citation is provided as a footnote on page one of each paper.

Printed in Baltimore, MD
November, 2011

Foreword

THIS COMPILATION OF THE *JOURNAL OF ASTM INTERNATIONAL (JAI)*, STP1533, on Surface and Dermal Sampling contains only the papers published in JAI that were presented at a symposium in San Antonio, TX, on 14-15 October 2010 and sponsored by ASTM Committee D22 on Air Quality and subcommittee D22.04 on Workplace Air Quality.

The Symposium Chairs and JAI Guest Editors are Michael Brisson, Savannah River Nuclear Solutions LLC, Aiken, SC and Kevin Ashley, Centers for Disease Control/National Institute for Occupational Safety and Health (NIOSH), Cincinnati, OH.

Contents

Asbestos

Pharmaceuticals

General Topics

Overview

This compilation represents the work of numerous authors at the ASTM International Symposium on Surface and Dermal Sampling, October 14-15, San Antonio, Texas, USA. This two-day symposium was sponsored by ASTM International Committee D22 on Air Quality and its Subcommittee D22.04 on Workplace Air Quality. The symposium was organized in cooperation with the American Industrial Hygiene Association (AIHA), the Beryllium Health and Safety Committee (BHSC), the U.S. Department of Energy (DOE), the U.S. Department of Housing and Urban Development (HUD), L'Institut de recherché Robert-Sauvé en santé et en sécurité du travail (IRSST), and the National Institute for Occupational Safety and Health (NIOSH) of the U.S. Centers for Disease Control and Prevention (CDC). Over thirty papers were presented at the symposium, and the papers that were submitted and accepted for publication appear in this volume.

The role of surface and dermal sampling to assess contamination levels, or to detect harmful agents, is growing. However, standard techniques for sampling of surfaces, including skin, are relatively few, and their development is hampered by limited data. The lack of harmonization in these techniques creates difficulties in comparing data from different studies. Agreement is needed on protocols for surface and dermal sampling and, to improve data defensibility, methods for sampling of surfaces, including skin, are in need of standardization. The symposium explored recent work that could aid in beginning the standards development process, and addressed challenges that need to be overcome for further standards development.

The symposium solicited presentations on the following topics (and related issues):

- Surface and dermal sampling protocols.
- Samplers and sample collection media.
- Target analytes — chemical, biological and radiation hazards, and dermal sensitizers.
- Application of surface and dermal monitoring techniques to real-world problems.
- Safety, health and risk assessment.
- Quality assurance and method performance.
- Policy issues relating to surface and dermal monitoring.

The targeted audience included a wide range of technical professionals such as industrial hygienists, chemists, biologists, health physicists, safety engineers, epidemiologists, medical personnel, and others having interest in surface or dermal sampling issues, or both.

The papers contained in this publication represent the commitment of ASTM International Committee D22 to providing timely and comprehensive information on advances in monitoring of toxic substances, exposure assessment, and standards development. Sections of the two-day symposium focused on the following themes: 1. Standardization Issues; 2. Dermal; 3. Lead; 4. Beryllium; 5. Asbestos; 6. Pharmaceuticals; and 7. General topics. Papers discussing sampling techniques, analytical measurement technologies, reference materials, standardization, occupational hygiene, decontamination methods, and quality assurance can be found in this compilation.

Standardization Issues

This section includes papers which summarize the currently available consensus standards for surface and dermal sampling and the need for additional standards, particularly in the area of dermal sampling. It also includes papers describing research activities intended to support standards development. Three of the papers that were given dealing with these issues are published in this section.

Dermal

This section includes papers dealing with aspects of addressing contaminants on skin, including sampling, removal, and adherence of contaminated materials to the skin. Two of the papers given in this topical area, both related to lead contamination, are published in this section.

Lead

This section includes papers dealing with sampling, sample preparation, analytical proficiency testing, and lead dust loadings on surfaces other than skin. Three of the papers that were given dealing with these issues are published in this section.

Beryllium

This section includes papers dealing with beryllium surface contamination in various industries and measurement of beryllium on surface wipe samples. Three of the papers given in this topical area are published in this section.

Asbestos

This section includes papers addressing the evaluation of samples collected from surfaces contaminated with asbestos. Two of the papers that were given dealing with these issues are published in this section.

Pharmaceuticals

This section includes papers dealing with occupational exposure to pharmaceutical substances, as well as spills and leakage of antibiotics on surfaces. Three of the papers given in this topical area are published in this section.

General Topics

This section includes papers addressing general topics such as investigations of outdoor environmental surface particulate; microbiological contamination on surfaces; assessments of properties contaminated with methamphetamine; and use of health-based screening levels to evaluate contamination on indoor surfaces. Four of the presented papers are published in this section.

We hope that readers of this publication will find it to be an informative and useful reference on surface and dermal sampling issues.

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Symposium Co-Chairs and Editors

Acknowledgments

The editors gratefully acknowledge the voluntary contributions of the numerous colleagues who served as peer reviewers of the manuscripts that were submitted for consideration for publication. Their efforts made the symposium and this compilation possible. Special thanks are extended to the following members of the symposium organizing committee, who helped to arrange the presentations and kindly served as session monitors:

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STANDARDIZATION ISSUES

Reprinted from JAI, Vol. 8, No. 6
doi:10.1520/JAI103678
Available online at www.astm.org/JAI

Kevin Ashley,[1] *Michael J. Brisson,*[2] *and Kenneth T. White*[3]

Review of Standards for Surface and Dermal Sampling†

ABSTRACT: This article summarizes the body of available standards for sampling of chemical and biological agents on workplace surfaces, including skin. These standards consist of voluntary consensus standards such as those promulgated by ASTM International, the International Organization for Standardization (ISO), and the European Committee for Standardization (CEN), as well as methods produced by U.S. Federal agencies such as the National Institute for Occupational Safety and Health (NIOSH) and the Occupational Safety and Health Administration (OSHA). Gaps in availabilities of standards are discussed along with activities underway to address needs in the field of occupational and environmental hygiene. In many cases, the available standards have been developed largely in response to regulatory requirements. For example, ASTM International standards, which describe requirements for wiping surfaces and methodologies for determining metals and metalloids such as lead and beryllium, were produced primarily in response to regulatory requirements for sampling settled dust for these elements in the United States. Methods for collection of asbestos samples, vacuum sampling, dry wipe sampling, and bulk sampling have also been promulgated. Standardized methods for non-metal contaminants and biological agents are more limited in availability. In particular, there is a lack of

Manuscript received December 6, 2010; accepted for publication April 28, 2011; published online July 2011.
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† This article was prepared by US government employees and contractors as part of their official duties and legally may not be copyrighted in the United States of America.

Cite as: Ashley, K., Brisson, M. J. and White, K. T., "Review of Standards for Surface and Dermal Sampling," *J. ASTM Intl.*, Vol. 8, No. 6. doi:10.1520/JAI103678.

standardized methodologies for dermal sampling and limited standard guidance on selection of appropriate surface sampling methods and data evaluation. Activities are currently ongoing within ASTM International and ISO to address some of the gaps, but additional activity is needed to address remaining requirements for consensus standards.

KEYWORDS: dermal exposure, sampling, skin, standard, surface, workplace

Introduction

Consistency in methods for sampling and analysis of chemical and biological agents from surfaces in occupational settings through standardization of methodologies is generally desired. However, incongruities in sampling and measurement practices often occur among those collecting and analyzing surface and dermal samples [1,2]. If sampling and analysis methods are not standardized, analytical results from different investigators, locations, and/or points in time might not be comparable. Variations in surface and dermal sampling practices are of special concern, since the greatest contribution to measurement uncertainty in the overall sampling and analysis process is ordinarily associated with sample collection. Efforts to control measurement uncertainty through method standardization have been realized for various hazardous agents in occupational settings. As a consequence, a number of standardized methods for surface sampling of hazardous substances in workplaces have been promulgated.

Several standardized protocols for surface and dermal sampling have been produced by governmental agencies in the United States, for example, by the Occupational Safety and Health Administration (OSHA) and the National Institute for Occupational Safety and Health (NIOSH). Voluntary consensus standards bodies such as ASTM International have also published a number of standardized protocols for the collection of surface samples. International voluntary consensus standards are considered by many to be the most technically sound protocols for use in their particular fields of application [3]. In part because consensus standards allow for stakeholder input and are recognized as having high credibility, the National Technology Transfer and Advancement Act (NTTAA; Public Law 104-113) was enacted in the United States in the mid-1990s. This law directs U.S. Federal agencies to: (a) rely on consensus standards in their guidelines, regulations, and activities and (b) participate in the development of relevant consensus standards. In accordance with the NTTAA, an earnest goal of experts from U.S. governmental agencies is to work with the private sector to provide a suite of consensus standards describing surface and dermal sampling methods for chemical and biological agents. As a starting point, many of the consensus standards under development are based initially on existing agency methods, guides, and procedures. Ultimately, it is intended that the use of consensus standards will enhance data comparability for surface and/or dermal samples obtained from different investigators, locations, and times.

Rationale for Surface Sampling (Non-Dermal)

Surface sampling results are one of the many sources of information regarding the health and safety conditions in workplaces or other locales. Information obtained from surface sampling should not be used to the exclusion of other information concerning potential chemical, radiation, and biological hazards; rather, surface sampling data should be used to augment data from other sources of contamination or exposure. For instance, additional sources of exposure information may include, as applicable: occupational air sampling; bioassay and biomonitoring results; clinical observations; quality and process control data; records of facility operations; visual inspections; and material balance studies. In an effort to address issues of this kind, an ASTM International consensus standard guide for surface sampling of metals has recently been published, which describes strategies for collecting surface samples for subsequent determination of metals and metalloids [4]. Many of the considerations outlined in this standard are applicable to other potentially hazardous agents besides metals, e.g., radiation hazards and biological agents.

Reasons for conduct of surface sampling are based on a number of general considerations. Drivers for sampling, that is, the purposes for carrying out a sampling campaign, normally fall into one of the following three areas: (a) evaluation of the potential health risk from the contaminant(s) or chemical agent, radiation hazard or biological species of concern; (b) hazard management, or evaluation of the source(s) of the contaminant or chemical species, radiation hazard or biological agent, extent of exposure area, and effectiveness of controls; and (c) hazard compliance, or evaluation for compliance with regulations or policies. Goals for the sampling campaign, which define how the generated data will be used, and a sampling strategy, should be clearly thought out and documented before any samples are collected. Of significant importance are the data quality objectives that define the minimum performance requirements for the collection and analysis of the samples. A related concern is the potential variability in surface contamination, which impacts the representativeness of collected samples. Sufficient numbers of samples and sampling areas of adequate size are required for defensible data to be obtained.

Besides workers' potential exposures, take-home contamination is also of particular concern [5]. Sampling and analysis of workers' clothing, vehicles, and home environments must be carried out to assess take-home contamination and to prevent exposures to family members.

The following are examples of purposes for surface sampling, as based on general considerations introduced above [4].

1. *Hazard identification and evaluation*—Estimation of the expected and/or or maximum concentrations of analyte(s) of interest in the workplace or other locale. The information obtained is used to evaluate risk, to recommend worker protection requirements, and to assess the probability of adverse health effects, including dermal responses such as contact dermatitis.
2. *Exposure assessment*—Collection of exposure data for when the existence of a health hazard is known or postulated. Assessment may

be focused on groups or populations of workers and/or family members, rather than on an individual worker. It requires, within limitations, the use of instrumentation and methods that offer the lowest available analytical reporting limits for the contaminant(s) of concern.

3. *Facility characterization*—Determination of the surface contamination levels of one or more analyte(s) of interest within a facility at an initial or baseline point, during or after process operations, or as part of facility decommissioning.
4. *Housekeeping*—Determination of the effectiveness of housekeeping actions. For example, wipe samples are often collected from cleaned surfaces to assess whether the cleaning procedure was effective in removing the contaminant(s) of interest.
5. *Selection of engineering controls*—Determination, for analyte(s) of interest that are not totally contained, of the collection or capture efficiencies of control devices necessary to bring specific contaminant concentrations below applicable limits at specific sampling locations; and for evaluation of the effectiveness of spill cleanup procedures.
6. *Evaluation of engineering controls*—Measurement of the quantities of analyte(s) of interest passing or escaping from a control device due to leaks, wear, damage, inadequate maintenance, overloading, or accidents.
7. *Evaluation of exposure pathways*—Measurements used as part of an evaluation of the potential contribution of (an) agent(s) of interest on surfaces to total worker, or workers' families', exposures. Assessment of the potential for take-home contamination might entail sampling of workers' clothing, shoes, and other items.
8. *Selection of personal protective equipment*—Determination of requirements for personal protective equipment in order for (a) worker(s) to inhabit a contaminated or potentially contaminated area for a specific period of time.
9. *Compliance with regulations and standards*—Measurements required to satisfy regulatory or legal requirements, to determine if exposures and/or contaminant surface concentrations in the workplace are below regulatory or established occupational exposure limits.
10. *Source identification*—Determination of the contribution from each of many potential sources to the presence of analyte(s) of interest, based on the unique characteristics of each of the agents of concern.
11. *Education and training*—Sampling, often accompanied with screening analysis, used to educate workers and managers in the importance of sound control practices such as engineering controls, personal protective equipment, and good housekeeping.
12. *Investigation of complaints*—Resolution of concerns expressed by workers, management, or other stakeholders.

Thus, in view of the above considerations, it is crucial to define the purpose(s) for collection of surface samples prior to conducting sampling. Defensibility of the data obtained is of primary concern. The use of standardized

protocols is more likely to enable potential data inter-comparisons and foster acceptance of the reported results.

Sampling for Assessment of Dermal Exposure

Considerations for the assessment of occupational dermal exposures have been proposed based on numerous scientific studies [6,7]. In view of this, the European Standards Committee (CEN) promulgated a standard technical report that outlines criteria for assessment of occupational dermal exposure [8]. A conceptual model of dermal exposure was outlined based on the extensive body of relevant scientific literature [6]. This model forms the basis of a protocol for choosing candidate measurement methods that can be used to assess dermal exposure contaminants and pathways. Dermal exposure assessment is often carried out through direct sampling from skin via wipe sampling, tape stripping, rinsing techniques, or in situ measurement methods, for example, Ref. [7]. Indirect dermal exposure assessment methods include, for example, patch sampling and sampling of clothing or gloves [6–8].

In general, four objectives for assessing dermal exposure can be highlighted [6]:

1. Research on adverse health effects of chemical exposures, including: (a) epidemiological investigations and risk assessment; (b) investigation of possible associations between skin exposure and adverse health effects; (c) development of exposure-response relationships for risk assessment; and (d) estimation of disease burden due to skin exposures.
2. Evaluation of exposure processes and pathways to assist in the development, implementation, and evaluation of exposure control measures or interventions.
3. Compliance, compensation claims, or litigation, if applicable.
4. Education and training, including intervention protocols that might include the use of screening techniques to aid in workers' understanding of their (and, potentially, their family members') exposure pathways.

Many of the considerations for carrying out dermal sampling mirror those outlined earlier for collection of non-dermal surface samples. Over 650 chemicals with "skin" notation have been identified [9], but the importance of dermal exposure is often underestimated or ignored.

Standardized Surface and Dermal Sampling Techniques

Representative substrates and sample media of interest that are applicable to surface and dermal sampling include, but are not necessarily limited to, the following:

- Hard/smooth/nonporous surfaces
- Soft/rough/porous substrates
- Fragile substrates
- Oily or coated surfaces
- Grossly contaminated surfaces
- Skin (exposed and/or protected)

- Clothing and personal protective equipment
- Patches, swabs, and tape
- Bulk materials, e.g., soils, deposited dust, and spilled materials

As an example, sampling techniques for metals have been promulgated that address sample collection from many of the above surface substrates. For maximum collection efficiency of metals (and excluding collection of bulk samples), "wet" sampling techniques using wipes are generally preferred [10,11]. However, there are situations where wet sampling of certain components and equipment are not desirable, and dry sampling techniques are required. For example, due to technical considerations, surfaces of certain materials and components must be protected against damage from the action of wetting agents and/or sample collection; hence, sampling methods that are less aggressive are sometimes required. For nonmetals, depending on the chemical or biological agent of concern, analogous considerations may be applicable.

In the case of surface sampling for metals, a hierarchy of sample collection methods is generally recommended [4]. At the outset, when it is determined that surface samples must be obtained, wet wipe sample collection methods are usually considered first. Such techniques are routinely applicable to smooth, hard, nonporous surfaces, and also to dermal sampling. Other sampling methods for various surfaces (including skin and clothing) consist of vacuuming methods, dry wiping protocols, tape stripping, rinsing techniques, and the use of swabs for collection of biological agents.

Table 1 summarizes standardized procedures for surface and dermal sampling that have been produced by OSHA [12] and NIOSH [13]; applications to sampling and analysis of metals and organics are exemplified. Dermal sampling by use of patch samples or rinsates has been described briefly in several OSHA and NIOSH methods (Table 1). Such techniques are presently planned for

TABLE 1—*OSHA [12] and NIOSH [13] procedures for sample collection from surfaces in occupational settings.*

Methods	Sampling Media/Device	Target Substrate(s) Sampled	Comments
OSHA ID-125G and ID-206	"Wet" or "dry" filter or wipe	Smooth surfaces, dermal samples	Alcohol wipes widely used; mainly applicable to metals
NIOSH 9100, 9101, 9102, 9105 and 9110	"Wet" wipes	Smooth surfaces, dermal samples	Metallic analytes: Pb, Cr(VI), Be, elements
OSHA Technical Manual (various)	Patch samples, hand rinsates	Dermal samples	Various protocols; also clothing, gloves, etc.; multiple analytes
NIOSH 3600, 3601, 9200, 9201, 9202 and 9205	Patch samples, hand rinsates	Dermal samples	Applicable to pesticides, metalworking fluids, etc.; may apply to other agents

further development as voluntary consensus standards within the ASTM International subcommittee on workplace exposure monitoring.

A number of ASTM International voluntary consensus standards pertaining to surface sample collection in workplace and building environments have been promulgated, and they are summarized in Table 2. ASTM standards have been published describing wet wipe sample collection of metals [14,15]. An ASTM surface tape stripping method has also been promulgated, and this technique is applicable to multiple analytes [16]. A tape stripping method for sampling of

TABLE 2—*ASTM International standards for sample collection from surfaces in workplaces and buildings.*

Standard	Sampling Media/Device	Target Substrate(s) Sampled	Comments
ASTM E1728 [14]	"Wet" wipe	Smooth surfaces	Applicable to Pb sampling; regulatory applications
ASTM D6966 [15]	"Wet" wipe	Smooth surfaces	Various wetting agents can be used; applicable to metals
ASTM E1216 [16]	Adhesive tape	Smooth surfaces	Poor collection efficiency for ultrafines; may damage fragile substrates; multiple analytes
ASTM D5438 [17]	Modified upright vacuum cleaner	Floors	Sampling from carpets; multiple analytes
ASTM D7144 [18]	Sampling cassette with collection nozzle	Rough, porous, uneven surfaces; fragile surfaces	"Micro-vacuum" dust sampling for metals; may be applicable to other agents
ASTM D7296 [19]	"Dry" wipe	Fragile surfaces	Applicable to beryllium only; special cases
ASTM E1792 [20]	Pb wipe specification	Smooth surfaces	Applicable to Pb sampling; may use for other metals; regulatory applications
ASTM D5756 [21]	"Micro-vacuum" sampler	General surfaces	Applicable to collection of asbestos fibers
ASTM D6480 [22]	Cloth: clean room wipe	Smooth surfaces	For collection of asbestos fibers
ASTM D6661 [23]	Solvent-wetted wipe	Smooth surfaces	Applicable to sampling of organic compounds
ASTM E2458 [24]	Swab sampler	General surfaces	Suspected biological agents in powders
ASTM D6333 [25]	Polyurethane foam roller	Floors	Applicable to pesticide residues

fungal spores is presently under development within ASTM International. When the surface to be sampled is rough or porous, and wet wipe sampling or tape stripping is deemed to be impractical, the use of vacuum collection methods is often considered in lieu of wiping or stripping techniques [17,18]. In rare cases where the surface to be sampled is energized, fragile, or reactive, and beryllium is the only analyte of interest, dry wipe sampling is an option for sample collection of this metal [19]. The use of wipes meeting the specifications of ASTM E1792 [20], while developed for wipe sampling materials for lead on surfaces, may be appropriate for sample collection of other metals. A related ASTM specification for wipe sampling materials for beryllium is presently under development. In addition, ASTM procedures for surface sampling of asbestos by vacuum sampling [21] or wiping, [22] wipe sampling of organic compounds [23], swab collection of biological agents [24], and collection of pesticide residues from floors [25] have been published (Table 2). Consensus surface sampling methods for lead [26], beryllium [27] and asbestos [28] have been developed largely in response to regulations in the United States. Besides the aforementioned ASTM standard surface sampling guide for metals [4], an analogous standard sampling guide for asbestos has also been developed [29].

Other ASTM standards relating to surface sampling and assessment of surface contamination have been developed to address applications in clean rooms and aerospace (Table 3) [30–33]. While these are specialized uses, there may be situations where the standards could be employed in contamination assessment in occupational and other environments.

The International Organization for Standardization (ISO) has promulgated a trio of international standards that address the measurement of surface contamination by radioactive materials [34–36]. These standards entail the measurement of radiation sources on surfaces of equipment and facilities, but they do not apply to the evaluation of radioactive contamination on skin and clothing. Methods for direct and indirect measurement of radionuclides collected from surfaces are described in these ISO standards.

TABLE 3—*ASTM International standard procedures for surface sampling in aerospace and clean room applications.*

Standard	Sampling Media/Device	Target Substrates Sampled	Comments
ASTM F303 [30]	Rinse method	Aerospace components	Collection of particulate matter for assessment of cleanliness
ASTM F51 [31]	Particle sizing instrument	Clean room garments	Evaluation of contamination from fibers and particles
ASTM E2088 [32]	“Witness” surface	Clean room surfaces	Measurement of particle deposition
ASTM F24 [33]	Optical particle counter	Electronic components	Assessment of surface contamination

Generic techniques for dermal sampling have been described in a European standard, EN TS 15729 [37]. This standard technical specification is a companion document to the aforementioned dermal sampling strategies technical report [8] that was developed by the same group. Currently, a draft ISO guidance document is under development that is based on these European standards and should soon be finalized.

Performance Data

In several cases, performance data have been published regarding collection efficiencies of some of the various surface sampling methods cited above. For instance, wet wipe sampling has been evaluated for the collection of lead oxide dust from smooth, hard surfaces, with sample collection efficiencies exceeding 75% routinely attained [38,39]. A minimum collection efficiency of 75 % has been specified for lead-containing settled dust that is sampled from smooth surfaces [20], and this criterion is generally applicable to other analytes. However, sampling from rough, porous or fragile surfaces cannot guarantee high collection recoveries, hence it is desired to harmonize the sampling procedures to the extent possible so as to enable reliable data comparisons.

In related work, a comparison of wet versus dry sampling was carried out on hard, smooth surfaces spiked with beryllium [40]. It was found that wet wipe sampling ordinarily results in a much higher collection efficiency (64 %–106 %) than does sample collection using dry wipes (14 %–43 %). In earlier studies, a comparison of wipe sampling methods for beryllium was carried out wherein dry, wet, and alcohol wipe methods were evaluated for their application in removing beryllium-containing dust from painted surfaces [41]. This investigation found alcohol to be most effective for removing beryllium dust from oily surfaces, while (not surprisingly) dry wipes were least effective for this purpose. These studies have served to provide necessary back-up data in support of standardized wipe sampling protocols for metals, e.g., ASTM D6966 [15].

The ASTM International high-volume vacuum collection method (ASTM D5438 [17]) for worn carpeted surfaces has been evaluated using reference material spikes, and good dust collection efficiencies (≈80 % and greater) have been reported for various types of carpets [42]. Previous investigations of this high-volume vacuum collection system on new carpets also reported effective collection (>75 %) of leaded dust from such substrates [43]. The more recently developed ASTM International low-air volume "micro-vacuum" collection method (ASTM D7144 [18]) was evaluated [44], and collection efficiencies from a variety of representative substrates were reported based on gravimetric analysis. Although recoveries were generally non-quantitative (<75 %), it was emphasized that standardization of the micro-vacuum sampling technique should ensure data comparability through harmonization of the sampling device and collection procedure. However, losses due to capture of significant amounts of material within the collection nozzles of the micro-vacuum samplers were reported [44]. This observation will hopefully lead to the design and development of improved samplers, where the collection inlet is incorporated into the

body of the sampler [45]. While removal of material from within the collection nozzles may be possible, in practice this is difficult to achieve.

In other research, assessment of dermal wipe sampling using different sample collection media has been carried out using lead as an analyte [46]. Most leaded dust (≈60 %) is recovered after sample collection with one wipe, although successive wiping increases overall dust removal (to ≈90 %) from workers' hands. Unfortunately to date, dermal sampling procedures have not been well standardized, and this has led to difficulties in evaluating and comparing data from a variety of different studies [47]. Data obtained by means of dermal sample collection techniques are often confounded by factors such as reactivity of the agent(s) of concern, analyte transport through skin, spatial variability of contaminant levels, and variance in the nature of skin surfaces [6,7]. Nevertheless, dermal sampling methods for chemical (e.g., pesticides, metalworking fluids) and biological agents (e.g., bacteria, viruses) need to be harmonized to the extent possible, and this remains an important area for further research and development.

Bulk Sample Collection

While methods for obtaining bulk samples are outside the scope of this article, they are briefly mentioned here since these techniques often complement surface and/or dermal sampling. An excellent source of information on bulk sampling methods for soils, solid waste, water, field equipment, etc., is the U.S. Environmental Protection Agency, which has published a comprehensive document [48] that covers issues such as: (a) Sampling strategies and design; (b) Sampling techniques, media, and equipment; (c) Standardized sampling procedures developed through voluntary consensus (notably ASTM International standards); and (d) Data quality considerations pertaining to sample collection, sample handling, and transport. A great many relevant ASTM International standards on collecting bulk samples have also appeared in compendium publications on environmental sampling [49,50]. Additional study is needed regarding when it is more appropriate to use bulk sampling in lieu of surface sampling. Performance data and guidelines are limited in this area.

Summary

The purpose of this article was to highlight the available standardized collection methods that are applicable to surface and dermal sampling. Within the arena of surface and dermal sample collection, our goal is to encourage the development of voluntary consensus standards in the areas of interest for which such standards are presently unavailable. Methods for surface sampling from smooth, hard surfaces are now reasonably well standardized, as evidenced by the availability of relevant international voluntary consensus standards. Additionally, vacuum sampling methods for collecting dust from rough, porous (and other) surfaces have also been standardized in the form of ASTM International procedures. General bulk sampling methods (not specific to dermal exposure

hazards) are also well standardized, and the use of voluntary consensus standards is encouraged. However, dermal sampling methods for chemical and biological agents require better harmonization and evaluation. Efforts are currently underway to fill these gaps.

Acknowledgments

The writers thank the members of ASTM International Committee D22 for their continued contributions and support. Drs. Aaron Sussell, Martha Waters, Bob Streicher, and Ken Brown kindly reviewed the draft manuscript. Mention of company names or products does not constitute endorsement by the Centers for Disease Control and Prevention, the U.S. Department of Energy, or their contractors. The contents and conclusions of this report are those of the authors and do not necessarily represent the views of the National Institute for Occupational Safety and Health.

References

[1] Lichtenwalner, C. P., "Evaluation of Wipe Sampling Procedures and Elemental Surface Contamination," *Am. Ind. Hyg. Assoc. J.*, Vol. 53, 1992, pp. 657–659.

[2] McArthur, B., "Dermal Measurement and Wipe Sampling Methods—A Review," *Appl. Occup. Environ. Hyg.*, Vol. 7, 1992, pp. 599–606.

[3] Office of Management and Budget (OMB), "Federal Participation in the Development and Use of Voluntary Consensus Standards and Conformity Assessment Activities," (OMB Circular No. A-119, Revised), 1998, www.whitehouse.gov/omb/circulars_a119#4 (Last accessed 30 Nov 2010).

[4] ASTM D7659, 2010, "Standard Guide for Strategies for Surface Sampling of Metals and Metalloids for Worker Protection," *Annual Book of ASTM Standards*, Vol. 11.07, ASTM International, West Conshohocken, PA.

[5] National Institute for Occupational Safety and Health (NIOSH), "Report to Congress on Workers' Home Contamination Study Conducted Under the Workers' Family Protection Act (29 U.S.C. 671a)," DHHS (NIOSH) Publ. No. 95-123. NIOSH: Cincinnati, OH, 1995, www.cdc.gov/niosh/95-123.html (Last accessed 30 Nov 2010).

[6] Schneider T., Vermeulen, R., Brouwer, D. H., Cherrie, J. W., Kromhout, H., and Fogh, C. L., "Conceptual Model for Assessment of Dermal Exposure," *Occup. Environ. Med.*, Vol. 56, 1999, pp. 765–773.

[7] Fenske, R. A., "Dermal Exposure Assessment Techniques," *Ann. Occup. Hyg.*, Vol. 37, 1993, pp. 687–706.

[8] EN TR 15728, 2006, "Workplace Exposure—Strategy for Evaluation of Dermal Exposure," European Committee for Standardization (CEN), Brussels.

[9] NIOSH, "NIOSH Pocket Guide to Chemical Hazards," NIOSH Publ. No. 2005-149. NIOSH: Cincinnati, OH, 2005, www.cdc.gov/niosh/npg (Last accessed 1 Nov 2010).

[10] Ashley, K., Braybrooke, G., Jahn, S. D., Brisson, M. J., and White, K. T., "Standardized Surface Dust Sampling Methods for Metals, With Emphasis on Beryllium," *J. Occup. Environ. Hyg.*, Vol. 6, 2009, pp. D97–D100.

[11] Ashley, K., Brisson, M. J., and Jahn, S. D., "Standardized Methods for Beryllium Sampling and Analysis—Availabilities and Needs," *J. ASTM Int.*, Vol. 2, No. 9, 2005, pp. 1–12.

[12] Occupational Safety and Health Administration (OSHA), "OSHA Sampling and Analytical Methods," OSHA Salt Lake Technical Center: Sandy, UT, 2003, www.osha.gov/dts/sltc/methods (Last accessed 24 Oct 2010).
[13] NIOSH, *NIOSH* Manual of Analytical Methods, 4th ed., (with supplements). NIOSH: Cincinnati, OH, 1994, www.cdc.gov/niosh/nmam (Last accessed 9 Nov 2010).
[14] ASTM E1728, 2003, "Standard Practice for Wipe Sampling of Surfaces for Subsequent Determination of Lead," *Annual Book of ASTM Standards*, Vol. 04.11, ASTM International, West Conshohocken, PA.
[15] ASTM D6966, 2008, "Standard Practice for Wipe Sampling of Surfaces for Subsequent Determination of Metals," *Annual Book of ASTM Standards*, Vol. 11.07, ASTM International, West Conshohocken, PA.
[16] ASTM E1216, 2006, "Standard Practice for Sampling for Particulate Contamination by Tape Lift," *Annual Book of ASTM Standards*, Vol. 15.03, ASTM International, West Conshohocken, PA.
[17] ASTM D5438, 2005 "Standard Practice for Collection of Floor Dust for Chemical Analysis," *Annual Book of ASTM Standards*, Vol. 11.07, ASTM International, West Conshohocken, PA.
[18] ASTM D7144, 2005, "Standard Practice for Collection of Surface Dust by Micro-vacuum Sampling for Subsequent Metals Determination," *Annual Book of ASTM Standards*, Vol. 11.07, ASTM International, West Conshohocken, PA.
[19] ASTM D7296, 2006, "Standard Practice for Collection of Settled Dust Samples using Dry Wipe Sampling Methods for Subsequent Determination of Beryllium and Compounds," *Annual Book of ASTM Standards*, Vol. 11.07, ASTM International, West Conshohocken, PA.
[20] ASTM E1792, 2003, "Standard Specification for Wipe Sampling Materials for Lead in Surface Dust," *Annual Book of ASTM Standards*, Vol. 04.11, ASTM International, West Conshohocken, PA.
[21] ASTM D5756, 2008, "Standard Test Method for Microvacuum Sampling and Indirect Analysis of Dust by Transmission Electron Microscopy for Asbestos Mass Surface Loading," *Annual Book of ASTM Standards*, Vol. 11.07, ASTM International, West Conshohocken, PA.
[22] ASTM D6480–05, (2010), "Standard Test Method for Wipe Sampling of Surfaces, Indirect Preparation, and Analysis for Asbestos Structure Number Concentration by Transmission Electron Microscopy," *Annual Book of ASTM Standards*, Vol. 11.07, ASTM International, West Conshohocken, PA.
[23] ASTM D6661, 2006, "Standard Practice for Field Collection of Organic Compounds from Surfaces Using Wipe Sampling," *Annual Book of ASTM Standards*, Vol. 11.04, ASTM International, West Conshohocken, PA.
[24] ASTM E2458, 2006, "Standard Practices for Bulk Sample Collection and Swab Sample Collection of Visible Powders Suspected of Being Biological Agents from Nonporous Surfaces," *Annual Book of ASTM Standards*, Vol. 15.08, ASTM International, West Conshohocken, PA.
[25] ASTM D6333, 2010, "Standard Practice for Collection of Pesticide Residues from Floors," *Annual Book of ASTM Standards*, Vol. 11.07, ASTM International, West Conshohocken, PA.
[26] Ashley, K., Schlecht, P. C., Song, R., Feng, A., Dewalt, G., and McKnight, M. E., "ASTM Sampling Methods and Analytical Validation for Lead in Paint, Dust, Soil, and Air," in *Sampling Environmental Media, ASTM STP 1282*; Morgan, J. H., Ed. ASTM International: West Conshohocken, PA, 1996, pp. 125–136.

[27] Rondeau, G. L., "Surface sampling," in *Beryllium—Environmental Analysis and Monitoring*, M. J. Brisson and A. A. Ekechukwu, Eds., Royal Society of Chemistry: Cambridge, U.K. 2009, Chap. 3.
[28] Beard, M. E., Millette, J. R., and Webber, J. S., "Developing ASTM Standards for Monitoring Asbestos," *ASTM Standard. News*, Vol. 32, No. 4, 2004, pp. 26–29.
[29] ASTM D7659, 2007, "Standard Guide for Evaluating Asbestos in Dust on Surfaces by Comparison Between Two Environments," *Annual Book of ASTM Standards*, Vol. 11.07, ASTM International, West Conshohocken, PA.
[30] ASTM F303, 2008, "Standard Practices for Sampling for Particles in Aerospace Fluids and Components," *Annual Book of ASTM Standards*, Vol. 15.03, ASTM International, West Conshohocken, PA.
[31] ASTM F51, 2007, "Standard Test Method for Sizing and Counting Particulate Contaminant In and On Clean Room Garments," *Annual Book of ASTM Standards*, Vol. 15.03, ASTM International, West Conshohocken, PA.
[32] ASTM E2088, 2006, "Standard Practice for Selecting, Preparing, Exposing, and Analyzing Witness Surfaces for Measuring Particle Deposition in Clean Rooms and Associated Controlled Environments," *Annual Book of ASTM Standards*, Vol. 15.03, ASTM International, West Conshohocken, PA.
[33] ASTM F24, 2009, "Standard Test Method for Measuring and Counting Particulate Contamination on Surfaces," *Annual Book of ASTM Standards*, Vol. 15.03, ASTM International, West Conshohocken, PA.
[34] ISO 7503-1, 1988, "Evaluation of Surface Contamination—Part 1: Beta-Emitters (Maximum Beta Energy Greater than 0.15 MeV) and Alpha-Emitters," International Organization for Standardization (ISO), Geneva.
[35] ISO 7503-2, 1988, "*Evaluation of Surface Contamination—Part 2: Tritium Surface Contamination*," ISO, Geneva.
[36] ISO 7503-3, 1996, "Evaluation of Surface Contamination—Part 3: Isomeric Transition and Electron Capture Emitters, Low Energy Beta-Emitters (Maximum Beta Energy Less than 0.15 MeV)," ISO, Geneva.
[37] EN TS 15729, 2006, "*Workplace Exposure—Measurement of Dermal Exposure, Principles and Methods*," CEN, Brussels.
[38] Chavalitnikul, C. and Levin, L., "A Laboratory Evaluation of Wipe Testing Based on Lead Oxide Surface Contamination," *Am. Ind. Hyg. Assoc. J.*, Vol. 45, 1984, pp. 311–317.
[39] U.S. Environmental Protection Agency (EPA), "Residential Sampling for Lead: Protocols for Dust and Soil Sampling," *Final Report No.* (EPA 747-R-95-001), EPA, Washington, D.C., 1995.
[40] Dufay, S. K. and Archuleta, M., "Comparison of Collection Efficiencies of Sampling Methods for Removable Beryllium Surface Contamination," *J. Environ. Monit.*, Vol. 8, 2006, pp. 630–633.
[41] Kerr, K., "Sampling Beryllium Surface Contamination Using Wet, Dry and Alcohol Wipe Methods," °M.S. thesis, Central Missouri State Warrensburg, MO, University, 2004.
[42] Svendsen, E., Thorne, P., O'Shaughnessy, P., Zimmerman, D., and Reynolds, S., "House Dust Collection Efficiency of the High Volume Small Surface Sampler on Worn Carpets," *J. Occup. Environ. Hyg.*, Vol. 3, 2006, pp. 333–341.
[43] EPA, "Laboratory Evaluation of Dust and Dust Lead Recoveries for Samplers and Vacuum Cleaners—Vol. 1: Objectives, Methods, and Results," *Report No.* (EPA 747-R-94-004A), EPA, Research Triangle Park, NC, 1995.

[44] Ashley, K., Applegate, G. T., Wise, T. J., Fernback, J. E., and Goldcamp, M. J., "Evaluation of a Standardized Micro-Vacuum Sampling Method for Collection of Surface Dust," *J. Occup. Environ. Hyg.*, Vol. 4, 2007, pp. 215–223.

[45] Creek, K. L., Whitney, G., and Ashley, K., "Vacuum Sampling Techniques for Industrial Hygienists, With Emphasis on Beryllium Dust Sampling," *J. Environ. Monit.*, Vol. 8, 2006, pp. 612–618.

[46] Boeniger, M., "A Comparison of Surface-Wipe Media for Sampling Lead on Hands," *J. Occup. Environ. Hyg.*, Vol. 3, 2006, pp. 428–434.

[47] Brouwer, D. H., Boeniger, M., and Van Hemmen, J., "Hand Wash and Manual Skin Wipes," *Ann. Occup. Hyg.*, Vol. 44, 2000, pp. 501–510.

[48] EPA, "RCRA Waste Sampling Draft Technical Guidance—Planning, Implementation and Assessment," *Report No.* (EPA 530-D-02-002), EPA, Washington, D.C, 2002.

[49] ASTM International, *ASTM Standards on Environmental Sampling*, 3rd ed., ASTM International, West Conshohocken, PA, 2006.

[50] ASTM International, *ASTM Standards on Environmental Site Characterization*, 3rd ed., ASTM International, West Conshohocken, PA, 2006.

Reprinted from JAI, Vol. 8, No. 6
doi:10.1520/JAI103654
Available online at www.astm.org/JAI

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Derivation of Health-Based Screening Levels for Evaluating Indoor Surface Contamination

ABSTRACT: This paper describes the development of U.S. Army Public Health Command (Provisional) Technical Guide 312, Health Risk Assessment Methods and Screening Levels for Evaluating Office Worker Exposures to Contaminants on Indoor Surfaces Using Surface Wipe Data. Surface sampling of indoor surfaces may be performed to determine whether a building is safe for re-entry following an event (for example, fire, pesticide application) or change in building use (for example, laboratory to administrative office). Surface wipe sampling results may be used to assess either the degree of contamination before cleanup or to determine whether post-abatement (also post-remediation, post-clearance) actions were effective. The U.S. Army Public Health Command (Provisional) Environmental Health Risk Assessment Program has conducted risk assessments based on surface wipe sampling data and, over time, has evolved the methodology into a technical guide. The technical guide addresses the need to develop an approach to characterize potential health risk to exposed populations using surface wipe sampling results and to provide surface wipe screening levels to facilitate initial assessments. The methodology provides a means to estimate office worker exposures from inhalation of resuspended particles, dermal contact, and incidental ingestion exposure routes. Potential chemical health risks from aggregated intakes are evaluated using conventional U.S. Environmental Protection Agency risk assessment methods.

Manuscript received November 30, 2010; accepted for publication April 29, 2011; published online July 2011.
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Symposium on Surface and Dermal Sampling on 14-15 October 2010 in San Antonio, TX; M. Brisson, K. Ashley, and J. Lesage, Guest Editors.

Cite as: Murnyak, G. and Chang, H.-Y., "Derivation of Health-Based Screening Levels for Evaluating Indoor Surface Contamination," *J. ASTM Intl.*, Vol. 8, No. 6. doi:10.1520/JAI103654.

KEYWORDS: health-based screening levels, surface wipe screening levels, exposure assessment, toxicity values

Nomenclature

ABS = absorption fraction
ADI_{derm} = average daily intake though skin
ADI_{ing} = average daily intake from ingestion
ADI_{inh} = average daily intake from inhalation
As = source area (meters squared $(m))^2$
AT = averaging time (days)
BW = body weight (kilograms (kg))
Cs = contaminant surface loading (milligrams per centimeter squared (mg/cm^2))
C_{wipe} = target surface wipe level
DL = detection limit
ED = exposure duration (year)
EF = exposure frequency (days/year)
ET = exposure time (hours/day)
EV_{derm} = event frequency for estimating the dermal dose (events/day)
$EVing$ = event frequency for estimating intake (events/day)
Fd = fraction exposed skin surface that actually contacts the surface (unitless)
F_f = fraction exposed skin area that contacts the mouth (unitless)
$FTsm$ = fraction of substance transferred from the skin to mouth (unitless)
f_{resp} = fraction respirable (unitless)
FT_{ss} = fraction transferred from surface to the skin (unitless)
i = subscript used to distinguish different parts of the exposed body (hand, forearm)
IR_{inh} = inhalation rate (m^3/h)
R = resuspension rate
SA = exposed skin surface area per event (cm^2/event)
$SWSL$ = surface wipe screening levels
V = room volume (meters cubed (m^3))
λ_{dep} = deposition loss rate (1/hour (h))
λ_a = air exchange rate (air changes per h)

Purpose

This paper describes the rationale and logic used by the U.S. Army Public Health Command (Provisional) (USAPHC (Prov)) formerly known as the U.S. Army Center for Health Promotion and Preventive Medicine (USACHPPM), to develop Technical Guide (TG) 312 [1]. The TG is designed to assist health risk assessors in interpreting indoor surface wipe samples in the context of human health risk. TG 312 provides a method for evaluating potential health risks to office workers from exposure to chemical substances on indoor work surfaces. The method may be used in two ways: (1) to

establish health-based surface wipe screening levels (SWSLs) as criteria to be compared with environmental wipe sample results or (2) to estimate cumulative health risks from exposure to chemical levels detected in wipe samples. Although TG 312 focuses on office worker exposures, the general method used to develop an exposure assessment may be adapted for other surface contaminant exposure scenarios by adjusting exposure factors used in the equations.

Need for Surface Limits

Surface sampling of indoor surfaces may be performed to determine whether a building is safe for re-entry following an event (for example, fire, pesticide application) or used for a different purpose (for example, industrial to office). Surface wipe sampling results may be used to assess either the degree of contamination before cleanup or to determine whether post-abatement (also post-remediation, post-clearance) actions were effective. Only a limited number of surface wipe standards or guidelines have been published [2]. Thus, there is a need to develop an approach to characterize potential health risks to exposed populations using surface wipe sampling results and to provide SWSLs to facilitate initial assessments.

Published Surface Wipe Standards or Recommended Limits

In contrast to airborne exposure limits, only a few standards or guidelines for surface levels of contaminants are published. This section summarizes some surface wipe levels that have been used to assess the hazards associated with surface contamination.

The U.S. Environmental Protection Agency (USEPA) provides cleanup levels for polychlorinated biphenyls (PCBs) in different media, which include nonporous surfaces [3]. The recommended PCB cleanup level for nonporous surfaces and high-occupancy use is ≤ 10 $\mu g/100$ cm^2 and applies to total PCB concentrations, not individual Aroclors [3].

The U.S. Department of Energy (USDOE) provides surface levels for beryllium to protect against chronic beryllium disease [4]. Two surface wipe levels are available. One applies to USDOE beryllium operations (3 $\mu g/100$ cm^2) and the other applies to beryllium-contaminated items before they are released to the general public (0.2 $\mu g/100$ cm^2) [5]. These limits were developed by USDOE based on reviews of historical work practice limits established by USDOE operational facilities using beryllium.

In response to the attacks on the World Trade Center (WTC), a task force was formed to select contaminants of concern and develop health-based benchmarks for exposure to indoor air and settled dust [6,7]. Health professionals used these benchmarks to determine if cleanup of residences were needed and, if so, to verify that the cleanup methods were effective. These benchmarks were developed based on both direct contact with indoor surfaces and incidental ingestion from hand-to-mouth transfer [6]. Inhalation of resuspended

particles was not included as air quality was monitored by taking actual air samples.

To address health concerns related to PCBs and 2,3,7,8 tetrachlorordibenzo-*p*-dioxin (TCDD) residues after a fire incident, Michaud et al., estimated risk-based re-entry surface wipe criteria for these substances [8]. Michaud et al., used a mass balance approach to propose re-entry surface level criteria applicable to full-time maintenance workers. These criteria were estimated using a lifetime risk level of 10^{-5}, applicable toxicological data, and professional judgement.

Evolution of TG 312

The methodology used in TG 312 evolved over time as USAPHC (Prov) health risk assessors evaluated surface wipe data from a variety of operations. The assessors prepared an interim report that provided surface screening levels for industrial scenarios [9]. This interim approach was updated by May and her colleagues who developed health-based screening levels using principles used by USEPA Regions III and IX for developing media-specific screening levels for cleanup activities. May developed screening levels for industrial and construction worker scenarios using three explosives as test substances [10]. The workers were presumed to potentially be exposed to surface contaminants from direct dermal contact, incidental ingestion from hand-to-mouth transfer, and from inhalation of resuspended particles. The methodology described by May, which was pending publication on September 11, 2001, was used to derive risk-based screening levels for PCBs, dioxins, and furans to assess potential health risks from exposure to surface contaminants at the Pentagon after the attack [11]. Gaborek et al., (2001) modified exposure parameters to reflect those of an office worker rather than an industrial or construction worker as in May et al., (2002) [10].

Technical Guide 312 uses a tiered approach that allows Army environmental professionals to assess potential health risks from exposure to surface contaminants. Surface wipe data can first be compared to SWSL values published in TG 312 if they are available. As a Tier 1 approach, the screening values are intended to have additional levels of conservatism for general applications. If surface wipe data exceed an SWSL, a more extensive assessment using the method described in TG 312 could be used for site-specific evaluations.

The methods used to measure surface contamination have been developed for nonporous surfaces and that limits interpretation of results to nonporous surfaces. Wood and concrete are common examples of porous surfaces while glass and vinyl are examples of nonporous surfaces.

Problem Definition

A fundamental problem with interpreting surface wipe sampling data in a health risk context is the difficulty in using surface wipe sampling data to

estimate chemical intake. To illustrate the problem, we can compare surface sampling scenarios to two simple exposure assessments for drinking water and fish consumption.

When chemical contamination of a drinking water source is a concern, a water sample can be submitted to the laboratory and the concentration of the chemical of concern is provided in milligrams of chemical per liter of water. With a known chemical concentration, we can estimate or measure the average liters of water consumed each day. Multiplying concentration by water consumption yields a straightforward estimate of daily intake of the chemical of concern.

Similarly, a scenario concerned with the health risk to a population associated with eating fish contaminated with a chemical can be estimated based on fish consumption. First, fish tissue samples need to be analyzed to estimate the concentration of chemical per kilogram of tissue. Next, we estimate the population's average fish tissue consumption per day. As with the drinking water example, multiplying fish consumption by the chemical concentration in fish tissue yields the estimate of chemical intake. These simple examples illustrate the general concept of estimating chemical intake for environmental health risk assessments.

In contrast, consider a surface wipe sampling scenario. Assume that we have the perfect sampling method and we know that the surface concentration of a chemical is exactly 50 μg/100 cm^2. The problem is that we have no direct calculation that helps estimate how the surface sampling data in this scenario yields a chemical intake. A major goal of TG 312 was to provide a methodology to estimate chemical intake under exposure conditions represented by surface contamination in a typical office scenario.

Due to different activity patterns and behavior, the specific exposure scenario is a vitally important consideration for estimating health risk. Visualize the previous surface wipe sampling dataset of 50 μg/100 cm^2 in a daycare setting with infants crawling and playing on the surface. Now contrast that daycare scenario with a locked machine room that one worker enters one time per year for a maintenance inspection. While we may not know the actual health risks, it is obvious that the same level of contamination can pose a different level of risk based on the use or activity scenarios.

Methodology

Basic USEPA Health Risk Approach

Typical USEPA risk assessments are based on the USEPA generic equation for calculating chemical intakes [12] (see Eq (1)). The generic equation is then adapted for the three routes of exposure designed to estimate potential intake from inhalation (resuspended particles), ingestion (associated with hand-to-mouth transmission), and dermal (contamination migration to the skin) routes of exposure

$$I = C \times \frac{CR \times EFD}{BW} \times \frac{1}{AT} \quad (1)$$

where:

I = intake (milligram/kilogram (mg/kg) body weight-day),
C = chemical concentration,
CR = contact rate (inhalation rate, ingestion rate, absorption rate),
EFD = exposure frequency and duration,
BW = body weight,
AT = averaging time.

Specific Enhancements of TG 312

Exposure Assessment

Exposure assessments can be completed using a variety of approaches [13]. Generally a health risk assessor combines information based on the activity patterns and behavior of the population of interest (exposure scenario), chemical concentrations in a medium, and the chemical's inherent hazard to estimate human health risk. Identifying the exposure scenario is a crucial first step to develop estimates of exposure and intake. The chemical intake estimate is compared to appropriate toxicity values to estimate health risk. The exposure scenario provides the assessor a picture of how the exposure is taking place and helps identify and organize the data needs and appropriate calculation methods. For TG 312, the exposure assumptions are provided only for an adult office worker population whose job functions involve the performance of typical office work tasks while the worker is seated at a desk. Literature searches and professional judgment were used to estimate behavioral and physical factors that impact worker exposure from surface contamination. When a worker performs an activity on a contaminated surface, some of the surface contaminants may be transferred to the skin. This transfer to skin can lead to both a dermal-absorbed dose and ingestion intake related to hand-to-mouth transfer. The inhalation exposure route in this method is limited to chemical intake as a result of activity that causes settled material to become resuspended in the air. Actual air sampling is recommended immediately following a primary release or when a continuous emission of airborne contamination is suspected.

Figures 1–3 illustrate the basic approach. Chemical intake is estimated for each of the three exposure routes, the estimated values are summed for a total intake, and then USEPA equations are used to calculate the potential health risks.

Dermal Route Example: Selection of Equation Parameter Values

The route-specific equations shown in Figs. 1–3 have been used to estimate exposure to indoor contaminants [10]. The TG 312 refines these equations by making them specific to comparison with surface wipes and more recently

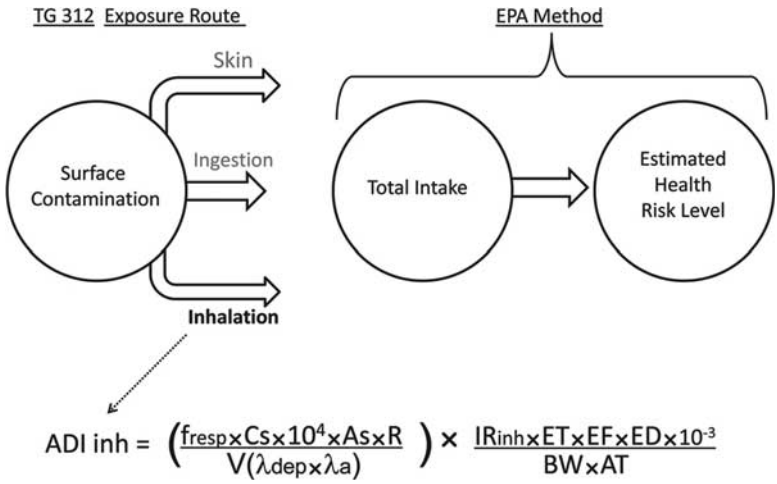

FIG. 1—*Inhalation exposure route.*

published data. However, the main difficulty was not in developing the mathematical models, but in selecting the appropriate parameter values. This selection is difficult due to a general lack of data, as well as standard testing protocol for generation of exposure data. These obstacles result from a limited understanding of the critical elements that affect a specific exposure and difficulty in designing test methods that reflect realistic exposures. This section focuses on the dermal exposure pathway to illustrate how recommended key parameter values were selected.

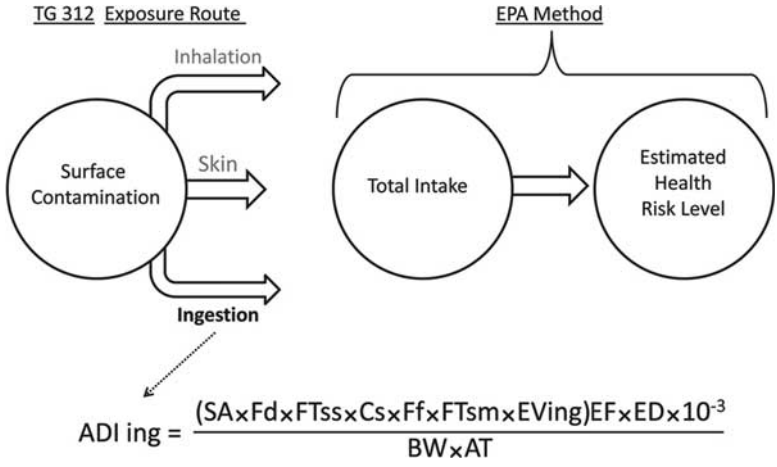

FIG. 2—*Incidental ingestion exposure route.*

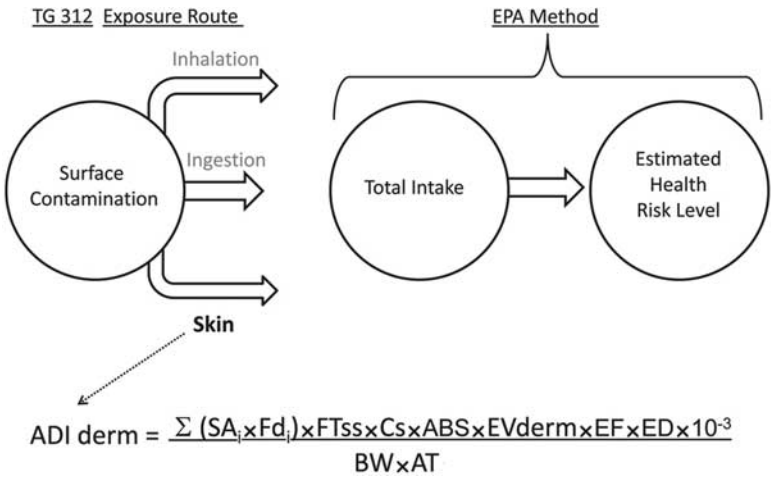

FIG. 3—*Dermal exposure route.*

To select appropriate values, it was necessary to identify relevant studies. For TG 312, this was facilitated by focusing on office worker exposures. Although there are less exposure data for office workers when compared to residential exposures, especially children, there is sufficient information to identify data gaps, limitations, and "test" the methods and concepts of TG 312. After relevant studies were identified, the next step was to determine whether the studies were comparable and, if not, understand any contradictions.

Figure 3 shows that for dermal exposure, the average daily intake is estimated by determining the potential dose per contact ($SA \times Fd \times FT_{SS} \times Cs$), averaged to an annual exposure based on the estimated number of contacts per year and normalized for the adult body weight. The potential dose is the dermal load on the skin available for absorption. This is a simplified way of expressing dermal exposure and probably overestimates actual intake because it does not consider losses from the skin, such as rubbing off the skin, but conservatively assumes that once on the skin all of the contaminant is available for absorption. Although TG 312 focuses on long-term exposure due to the lack of acute dermal toxicity data, the potential dose could be used to estimate short-term exposures if acute toxicity data is available. The paragraphs below discuss the parameters SA, Fd, and FT_{SS} in greater detail.

Surface Area (SA)—Surface area estimates the amount of exposed skin area in contact with contaminated surfaces. This takes into consideration common office attires and "surface contact traits" to account for the fact that not all of the exposed SA would come in contact with a contaminated surface. For example, an office worker may rest an arm on the tabletop but would not normally roll the bare arm around the tabletop. In addition, although some compounds

may penetrate clothing, only uncovered body parts were considered because TG 312 focuses on exposure to residues and not bulk chemicals (for example, spills). A case study prepared for the Office of Pesticide Non-Dietary Subcommittee, referred to proprietary data citing a 100-fold difference between transfer factors measured for single-layer clothing and for uncovered body parts [14]. This supports the decision to omit covered body parts because the amount of chemical that penetrates clothing, especially at residual levels, is not expected to significantly increase the overall exposure.

For estimating long-term exposures, it was determined that the forearms and hands are the most frequent body parts to come in contact with contaminated surfaces. To account for "surface contact traits," two-thirds of the forearm and only the palmar sides of the hands were considered to be most frequently in contact with contaminated surfaces. The two-thirds factor is an arbitrary adjustment factor that considers the underside of the forearm and the "spread-out" effect of the arm when it is laid on a flat surface. It was further assumed that both forearms and hands are simultaneously in contact with the surface. For acute exposures, it may be necessary to reconsider the total exposed SA to account for larger SAs such as when an office worker wears a skirt and part of the leg is exposed to the chair surface. The uncertainty that the undersides of upper legs have not been included in the SA estimate for long-term exposures is offset by other conservative assumptions such as the simultaneous contact of all exposed areas and the assumption that the total loading on the skin surface is available for uptake through the skin.

Forearm SAs were obtained from the USEPAs Exposure Factors Handbook (EFH). Since SA and body weight are codependent variables, 50th percentile values were selected to correlate with the mean adult body weight of 70 kg commonly used in risk assessments. Literature values of adult palmar surface areas have been reported in several studies [15,16]. These values are preferred over whole hand SAs reported in the EFH, which would require assigning an adjustment factor to obtain palmar surface areas. There is not much variation in measured hand surface areas other than the limitation that they are only for male subjects. However, this is not an issue for TG 312. To provide conservative screening values, male SAs were used because they are, in general, higher than female skin SAs.

Fraction of Skin Surface Area That Actually Contacts the Surface (Fd)—This parameter modifies the exposed skin SA to reflect the area that actually touches the surface. Even though TG 312 was simplified by focusing only on nonporous, flat surfaces, trying to define a single value for deriving screening values was difficult because experimental studies indicate that Fd is affected by different factors. These factors include particle size, contaminant surface loading, number of repeated contacts, pressure applied, and surface type [16–18]. Table 1 summarizes values of Fd reported for hands in contact with smooth surfaces. The table does not include adjustments for potential false negatives associated with the video imaging system employed by Brouwer to measure exposed area. The authors reported it to be reasonable to expect the percent of exposed area after

TABLE 1—*Summary of experimental data on actual hand area exposed (smooth surfaces).*

Study	Surface Loading ($\mu g/cm^2$)	Measured Area (cm^2)	Percent of Exposed Area (%)	Number of Repetitive Contacts	Pressure Applied
Brouwer et al., (1999)	6	7.0	4	1	0.005 kg/cm^2
	177	26.6	16	1	
	177	61.9	39	12	
Rodes et al., (2001)	None	Not reported	37.2 (mean, n=4)	1	5.4 kg
		61.1	35 (n=1)	1	
Zainudin (2005)[a]	1830[b]	Not reported	10	1	Not reported
			34	6	

[a]Data based on contact with smooth surface and tool (pliers or screwdriver).
[b]Estimated from data presented in Zainudin (550 mg solid and smooth area of 20 cm × 15 cm).

12 repeated contacts to range between 40% and 54% after correcting for false negatives.

At the time of the development of TG 312, no office activity-specific data were found; therefore, results from repeated contact studies were used to estimate Fd for typical office activities. After reviewing reported data and using professional judgment, a value of 0.30 was used to account for the palmar side of the hand that actually comes in contact with the surface. This value was rounded down from 34% (0.34) for six repeated contacts reported by Zainudin and Semple. Some factors were considered when rounding down this value. First, most surface contacts in the office do not involve carefully applied hand presses to the surface. While occasional hand presses may occur, since the TG 312 focuses on long-term exposures, it was more appropriate to consider the most likely daily contacts and not occasional contacts resulting in high exposures. Second, TG 312 focuses on contacts with smooth, nonporous surfaces. Since Zainudin's results include subjects grasping hand tools, those values probably overestimate Fd for casual contacts with surfaces in an office environment. Finally, unless the contaminated surface is loaded with a large amount of particles, the maximum Fd is probably much less than 50% for casual contacts.

Experimental values that could be used to estimate Fd are limited to hand data. Therefore, they would not be applicable to forearms because the hand has creases and joints, but the forearm is comparatively smoother. Since the forearm is smooth, it was assumed that 100% of the forearm in contact with the surface actually contacts the surface. The differing Fd terms for each body part exposed explain why the equation shown in Fig. 3 includes a summation of SA × Fd. Whether the default Fd values used in TG 312 could be "borrowed" to assess exposures to porous surfaces is not known since there are no similar data reported for porous surfaces.

TABLE 2—*Summary of factors that affect FT_{ss} values.*

Factor	Comments/Supporting Studies
Method of measuring amount transferred to the skin and experimental design	• Currently, there is no one protocol for measuring the transfer of surface residue to the skin. This makes comparison between studies difficult. • Use of a pre-cleaned versus a "dirty" test surface may have an effect on the amount transferred to the skin.
Type of solvent used to recover amount transferred to the skin	• For hand-wash techniques, use of solvents improves surface contaminant recovered from the hands. However, an ideal solvent for one contaminant may not be the ideal choice for another. • Fenske and Lu (1994) showed time between contact and hand wash did not affect chlorpyrifos recovery when ethanol was used but did affect chlorpyrifos recovery when isopropanol/water was used [19].
Skin moisture level and type	• Dampness of the skin affects the amount transferred. Damp skin picks up more residue/particles from the surface than dry skin. • Studies suggest the type of natural solvents is not a significant factor in contaminant transfer from the surface to the skin. • Transferability is also affected by contact surface moisture. Transferability is greater for wet surfaces [20].
Intensity of surface contact (e.g., rubbing, pressure applied)	• Smudging, amount of pressure applied to the surface during contact, and the number of successive contacts may increase transfer. Contact with a clean surface may result in removal of contaminants already transferred to the hands [17].
Contaminant surface loading	• Increasing surface loading results in decreasing transfer efficiencies, which indicates a specific skin surface area is not a limitless receptor.
Contaminant type/formulation	• Transfer efficiency will be affected by the type of residue (e.g., bound to dust, dried on surface) on the surface. For pesticides, the formulation (e.g., wettable powder, flowable) may also affect transfer efficiency [19]. • Some compounds transfer better to the skin. Percent pyrethrin transferred was about two times higher than chlorpyrifos or piperonyl butoxide [21]. Ramwell postulated transferability is affected more by a surface contaminant's octanol–water partition coefficient and less by its solubility [20]. • Smaller particles are more likely to stick to the skin because of strong adhesive forces [16]. • One study involving microorganisms showed positively charged bacteria transferred more readily to other surfaces than gram-negative bacteria or viruses [22]. This study suggests transfer efficiency may be affected by the charge of the surface contaminant.

TABLE 2—*Continued*

Factor	Comments/Supporting Studies
Surface contact time	• No significant change in transfer efficiency was observed between contact durations ranging from 3 to 30 s [17]. The effect of a wider time range on the transfer efficiency is unclear. McArthur and Lees (1995) noted significant differences in mass transferred from a smooth surface for contact times ranging from 5 to 20 min. However, McArthur used oil as a test substance and a porous medium to collect oil from the surface. It is unknown whether McArthur's observations could be applied to transfer of particles to the skin [23].

Fraction Transferred From the Surface to the Skin (FTss)—The FT_{ss} estimates the amount of contaminant transferred to the skin upon contact with the surface. Depending on the objective of a study, FT_{ss} is sometimes referred to as "removal efficiency." For example, studies designed to compare different surface sampling methods by measuring the amount of residue "removed" by the hands often use the term "removal efficiency" to describe the amount of residue on the hands. Results from these studies could be used to estimate values of FT_{ss}. Using values from studies with different objectives and often different measurement techniques make comparison of results difficult at times. However, even if a direct comparison of the results cannot be made, these studies are all critical to understanding the factors that affect FT_{ss}.

A detailed discussion of relevant studies used to identify factors that affect the amount of substance transferred to the skin is provided in TG 312 and will not be repeated here. However, Table 2 summarizes major factors identified from these key studies. These factors were obtained from two general types of studies. One type involves direct measurement of contaminant mass on the hands after contact with the surface. Most of these studies are limited to measurements of residues after pesticide application. The second type of data concern is measurement of particles such as dust or ground-up Tinopal®[3]. Results from these studies are typically reported as surface loading (mg particle/surface area) and require some assumption to relate the dust loading to the amount of contaminant transferred to the skin surface. The simplest approach is to assume the contaminant is evenly distributed so the fraction of contaminant transferred is the same as the fraction of particles transferred to the skin.

Reported amounts of substance transferred from surface to skin range widely, varying from less than 1% to over 100%. High transfer efficiencies were reported for experimental conditions involving vigorous rubbing or use of solvents, which removed more contaminants from the surface than from normal

[3]Tinopal is a registered trademark of Ciba-Geigy Corporation, 444 Saw Mill River Road, Ardsely, NY 10502.

surface contacts. Therefore, most of the reported experimental data could not be used to estimate an FT_{ss} value for deriving SWSLs. For TG 312, a deterministic value of 6.3% was used to estimate the amount of contaminant transferred to the skin from surface contacts during normal office activities such as working at a desk. This value is based on results reported by Brouwer (1999) for six repeated contacts at a low surface loading of 6 $\mu m/cm^2$. Repeated contact studies were preferred over single contact studies because experimental data have shown that the amount transferred from the surface to hands increases with consecutive contacts [16,17]. However, studies that exaggerate repeated contacts, such as repeated single fingertip presses used by Rodes et al., (2001), were not considered. Repeated fingertip contacts are easy to control experimental factors so that changes in transfer rate can be observed, but would overestimate actual exposure from normal contact with surfaces.

Developing SWSLs

The SWSLs were derived using the basic USEPA equation for estimating intake (Eq (1)) and back-calculating to a surface concentration. Intakes from all three exposure pathways are aggregated to obtain a single surface concentration for each substance. The TG 312 uses the following steps to derive a substance-specific SWSL:

- Step A Calculate Cs based on carcinogenic effects;
- Step B Calculate Cs based on noncancer effects;
- Step C Compare values calculated from Steps A and B and select lower value;
- Step D Calculate target surface wipe level (C_{wipe}) using value selected in Step C;
- Step E Compare C_{wipe} to the analytical detection limit (DL) of the substance. If C_{wipe} is lower than the DL, set DL as screening level.

Step A, if applicable, is calculated by rearranging the three equations in Figs. 1–3 and multiplying each route-specific intake by the respective cancer slope factor. Surface concentrations based on systemic effects are calculated the same way, except each route-specific intake is divided by the respective reference dose

TABLE 3—*Comparison of TG 312 screening values to other published values.*

Substance	Source	Safe Level ($\mu g/100\ cm^2$)
Beryllium	USDOE	3 and 0.2
	TG 312	4.7
PCB	Toxic Substances Control Act (TSCA) USEPA	10
	TG 312	1.60 and 9.04
Michaud et al., [8]	7.5	
2,3,7,8 TCDD	USEPA WTC	0.00002
	TG 312	0.0000354
	Michaud et al., [8]	0.00125

or reference concentration. Depending on risk management objectives, different target levels for carcinogenic and noncancer effects could be considered. For TG 312, a target level of 1E-06 is used to calculate surface concentrations for carcinogenic effects. This target level corresponds to an excess lifetime cancer risk of 1 in 1 million people when compared to background exposures. The potential for noncancer effects is evaluated by comparing the daily intake to a threshold effects value, and a level of 1 is typically used as the target level. Therefore, for TG 312, a target level of 1 is used for noncancer effects.

A review of existing literature and consultation with a USAPHC (Prov) analytical chemist with extensive experience in surface sampling suggest that surface sampling methods are still not well characterized. Therefore, in Step D, adjustment factors of 0.50 for organics and 0.75 for metals were added to account for sampling and analytical variabilities. For example, for metals, the surface concentration calculated from Step C is multiplied by 0.75, resulting in a lower value. A higher sampling efficiency was used for metals because there is less variability in wipe sampling procedure and analytical methods. Since SWSLs are intended to be screening levels for general application, it is justifiable to include an additional level of conservatism. These adjustment factors could be removed or modified depending on information available to the evaluator or risk management decisions. When compared with other published screening levels (Table 3), the SWSLs do not appear to be overly conservative or unreasonable. The value for 2,3,7,8-TCDD is higher than the value used for the WTC evaluation. This is reasonable because the values for WTC are intended for residential exposures while the factors considered for derivation of the SWSLs in TG 312 are intended for office environments. More detailed discussions of the different sampling efficiencies reported in the literature as well as a summary of difference sampling methods used by various agencies are provided in TG 312.

Final SWSLs are adopted only after comparison with the substance's analytical DL (Step E). The SWSLs based on the substance's DL are highlighted so that the user knows that the SWSL is not a health-based value and further evaluation may be necessary. For example, either the analytical DL could be lowered or the individual parameter values used to estimate exposure and intake could be modified based on site-specific information.

Table 3 provides a comparison of the SWSL values calculated using the TG 312 method compared to some published surface wipe criteria. The table helps to support the methodology because the calculated TG 312 SWSLs are comparable to these published values.

Limitations

Potential sources of uncertainty can be associated with the parameters selected for the risk assessment model and the inherent uncertainty with the model itself. Some sources of uncertainty may be reduced by further data collection or refinement of model parameters. Collecting more environmental samples can better characterize the environmental conditions, and further literature searches may provide better estimates for model parameters.

Calculated SWSLs are sensitive to assumptions and default input variables. Assumptions, uncertainty, and limitations are unavoidable because of gaps in our knowledge concerning exposure conditions and toxic responses. The screening level values in this TG have the inherent scientific uncertainty associated with the health-based reference dose and the unit risk values used in the calculations and as described in the USEPA integrated risk information system [24]. The exposure assessment used in this TG to develop the screening values also has uncertainties related to characterizing the three routes of exposures used to develop the exposure assessment.

Health professionals applying this screening level methodology need to consider the potential for multiple chemicals within acceptable risk ranges having an additive impact on total risk. Screening level values are developed for single chemicals and are evaluated against their chemical-specific screening level value. It is possible that all individual chemicals present may be below their specific screening level values, yet the sum of the risk may exceed an acceptable value. Multiple chemical exposure scenarios may warrant more detailed risk assessments if there is a question.

Conclusion

Surface wipe samples collected from potentially contaminated indoor surfaces are the tool used by environmental and occupational health practitioners to help evaluate health hazards from chemical exposures. However, the utility of surface wipe samples have been limited by the lack of generally accepted standards or health criteria regarding surface contamination limits. As a result, surface wipe results are often used as a means to detect or not to detect a chemical of concern on an indoor surface rather than to actually inform a quantitative health risk assessment.

The USAPHC (Prov) environmental health risk assessors wrote TG 312 to document an exposure assessment methodology that provides a means to calculate a chemical intake from surface wipe sample results. Once the surface wipe results are expressed as a chemical intake, standard USEPA risk assessment equations are applied to calculate an estimated health risk. Additionally, TG 312 presents a methodology that allows Army practitioners to select an acceptable level of risk and uses these same equations to calculate conservative SWSLs. These SWSLs can be used by army environmental and occupational health practitioners to identify and prioritize chemicals of concern and conduct site-specific evaluations as required. While the specific exposure scenario developed in TG 312 is for general office work, experienced health risk assessors may tailor the exposure parameters to match a different exposure scenario and apply the same basic equations.

References

[1] USACHPPM, *Technical Guide 312, Health Risk Assessment Methods and Screening Levels for Evaluating Office Worker Exposures to Contaminants on Indoor Surfaces*

Using Surface Wipe Data, USACHPPM, Aberdeen Proving Ground, MD, 2009, http://phc.amedd.army.mil/PHC Resource Library/TG 312 (Health Risk Assessment Methods and Screening Levels for Evaluating Office Worker Exposures.pdf

[2] Ness, S. A., *Surface and Dermal Monitoring for Toxic Exposures*, Wiley, Hoboken, NJ, 1994, p. 128.

[3] USEPA, "Polychlorinated Biphenyl (PCB) Site Revitalization Guidance Under the Toxic Substances Control Act (TSCA)," USEPA Report No. OPPT-2004-0123, Office of Pollution Prevention and Toxics, Washington, DC, 2005, Available from: http://www.epa.gov/osw/hazard/tsd/pcbs/pubs/pcb-guid3-06.pdf.

[4] "Chronic Beryllium Disease Prevention Program; Final Rule," 1999, Federal Register, 64 (No. 235; Dec. 8), 68854–68914, Available from: http://www.gpoaccess.gov/fr/search.html.

[5] Code of Federal Regulations, Title 10, Chapter 3, Subchapter 1, Part 850, Subpart c.

[6] Contaminants of Potential Concern (COPC) Committee of the World Trade Center (WTC) Indoor Air Work Group (IAWG), *World Trade Center Indoor Environment Assessment: Selecting Contaminants of Potential Concern and Setting Health-Based Benchmarks*, COPC Committee of the WTC IAWG, New York, NY, 2003, Available from: http://www.epa.gov/wtc/copc_benchmark.pdf.

[7] USEPA, "World Trade Center Residential Dust Cleanup Program: Final Report," *Prepared by USEPA Region 2*, New York City Response and Recovery Operations, New York, NY, 2005.

[8] Michaud, J. M., Huntley, S. L., Sherer, R. A., Gray, M. N., and Paustenbach, D. J., "PCB and Dioxin Re-Entry Criteria for Building Surfaces and Air," *J. Expo. Anal. Environ. Epidemiol.*, Vol. 4, No. 2, 1994, pp. 197–227.

[9] USACHPPM, "Derivation of Wipe Surface Screening Levels for Environmental Chemicals: Industrial Scenarios," USACHPPM Project No. 39-EJ-1157-99: Interim Report, USACHPPM, Aberdeen Proving Ground, MD, 1999.

[10] May, L. M., Gaborek, B., Pitrat, T., and Peters, L., "Derivation of Risk Based Wipe Surface Screening Levels for Industrial Scenarios," *Sci. Total Environ.*, Vol. 288, No. 1–2, 2002, pp. 65–80.

[11] Gaborek, B. J., Mullikin, J. M., Pitrat, A. T., Cummings, L., and May, L. M., "Pentagon Surface Wipe Sampling Health Risk Assessment," *Toxicol. Ind. Health*, Vol. 17, No. 5–10, 2001, pp. 254–261.

[12] USEPA, "Risk Assessment Guidance for Superfund, Volume 1: Human Health Evaluation Manual (Part A)," *EPA/540/1-89/002*, USEPA, Office of Emergency and Remedial Response, Washington, DC, 1989, Available from: http://www.epa.gov/oswer/riskassessment/ragsa/pdf/rags-vol1-pta_complete.pdf.

[13] USEPA, "Guidelines for Exposure Assessment," *EPA/600/Z-92/001*, USEPA, Washington, DC, 1992, Available from: http://rais.ornl.gov/homepage/GUIDELINES_EXPOSURE_ASSESSMENT.pdf.

[14] USEPA, *REx: Residential Exposure Assessment Generic Methods, Case Study: Lawn Care Products*, OP Case Study Group, Non-Dietary Subcommittee, Washington, DC, 1999.

[15] Edwards, R. D. and Lioy, P. J., "The EL Sampler: A Press Sampler for the Quantitative Estimation of Dermal Exposure to Pesticides in Housedust," *J. Expo. Anal. Environ. Epidemiol.*, Vol. 9, 1999, pp. 521–529.

[16] Rodes, C. E., Newsome, J. R., Vanderpool, R. W., Antley, J. T., and Lewis, R. G., "Experimental Methodologies and Preliminary Transfer Factor Data for Estimation of Dermal Exposures to Particles," *J. Expo. Anal. Environ. Epidemiol.*, Vol. 11, No. 2, 2001, pp. 123–139.

[17] Brouwer, D. H., Kroese, R., and Van Hemmen, J. J., "Transfer of Contaminants From Surface to Hands: Experimental Assessment of Linearity of the Exposure Process, Adherence to the Skin, and Area Exposed During Fixed Pressure and Repeated Contact With Surfaces Contaminated With a Powder," *Appl. Occup. Environ. Hyg.*, Vol. 14, No. 4, 1999, pp. 231–239.

[18] Zainudin, A. and Semple, S., "Determination of the Factors Influencing Transfer of Chemicals From the Work Environment to the Skin and Hence From Hands to the Peri-Oral Area," *First International Occupational Hygiene Association (IOHA) International Scientific Conference in Africa*, Pilanesberg National Park, South Africa, Sept. 19–23, 2005.

[19] Fenske, R. A., and Lu, C., "Determination of Handwash Removal Efficiency: Incomplete Removal of the Pesticide Chlorpyrifos From Skin by Standard Handwash Techniques," *Am. Ind. Hyg. Assoc. J.*, Vol. 55, No. 5, 1994, pp. 425–432.

[20] Ramwell, C. T., Johnson, P. D., and Corns, H., "Transferability of Six Pesticides From Agricultural Sprayer Surfaces," *Ann. Occup. Hyg.*, Vol. 50, No. 3, 2006, pp. 323–329.

[21] Clothier, J. M., "Dermal Transfer Efficiency of Pesticides From New, Vinyl Sheet Flooring to Dry and Wetted Palms," *EPA/600/R-00/029*, USEPA: Office of Research and Development (Contract No. 68-DO-0049), Research Triangle Park, NC, 2000, Available from: http://www.healthyhousing.org/clearinghouse/docs/Article0022.pdf.

[22] Rusin, P, Maxwell, S., and Gerba, C., "Comparative Surface-to-Hand Fingertip-to-Mouth Transfer Efficiency of Gram-Positive Bacteria, Gram-Negative Bacteria, and Phage," *J. Appl. Microbiol.*, Vol. 93, 2002, pp. 585–592.

[23] McArthur, B. R., and Lees, P. S. J., "Effect of Contact Time and Contact Pressure on the Transfer of Oil to Surface Sample Media," *Appl. Occup. Environ. Hyg.* Vol. 10, No. 1, 1995, pp. 23–27.

[24] USEPA, "Integrated Risk Information System," USEPA: Office of Research and Development, National Center for Environmental Assessment, Washington, DC, 2008, Available from: http://www.epa.gov/iriswebp/iris/ (accessed April 2008).

Reprinted from JAI, Vol. 8, No. 6
doi:10.1520/JAI103514
Available online at www.astm.org/JAI

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Indoor Allergen Surface Sampling Methods and Standards: A Review of the Theory and the Practice*

ABSTRACT: Understanding the relationship among allergies, asthma, and indoor air allergen triggers increasingly require use of standardized and evidence based exposure assessment methods. Exposure assessment for common indoor allergens, however, is often limited by use of surface in place of air sampling for a variety of reasons. Chief among them is that many indoor allergens settle to the ground quickly after being released. Another reason for the wide-spread use of surface over air sampling for indoor air allergens is that air sampling is typically performed over a limited time frame and plausibly cannot capture a composite of exposure that a surface sample can. Protocols have been developed to collect surface borne allergen in several US wide-national studies, and while these samplers may provide an adequate means to correlate composite dust mass or allergens to some specific health outcomes, there is still a lack of evidence to support their adoption on the basis of several criteria which stem from theoretical and evidence based considerations. To become more valuable tools to risk assessment, epidemiological studies, and environmental intervention, surface sampling methods should be constructed using basic principles of particle behavior on surfaces as well as in the air. Criteria need to be developed from these principles and studies that address these criteria should be used or developed to enable the creation of performance-based standards. This paper is a review of the literature, which highlights surface sampling methods that have taken this theory- or evidence-based approach. After the review, a discussion is then developed

Manuscript received October 18, 2010; accepted for publication April 29, 2011; published online July 2011.

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* The authors would like to dedicate this paper to the memory of John W. Roberts, a pioneer in "Healthy Home" efforts and environmental sampling and control.

Cite as: Lewis, R. D. and Chen, D., "Indoor Allergen Surface Sampling Methods and Standards: A Review of the Theory and the Practice," *J. ASTM Intl.*, Vol. 8, No. 6. doi:10.1520/JAI103514.

on the current state-of-the-art surface sampling for indoor allergens. Recommendations are suggested for both future theoretical or empirical work necessary for devising performance standards for sampling and collecting surface borne-allergens.

KEYWORDS: surface, sampling, allergen, dust, resuspension, dust mite, mold, bacteria

Introduction

More than fifteen years ago, a committee of the Institute of Medicine was formed to address the subject of Indoor Allergens. There were several findings of the committee including recommendations to conduct further research to: standardize methods of collecting and analyzing indoor allergen samples to facilitate comparative and collaborative studies, quantitate the relationship of allergens in reservoir (surface) dust to airborne or aerosolized allergens, develop exposure metrics analogous to time-weighted averages or permissible exposure limits for allergens, and establish effective mechanisms for medical professionals to acquire assessments of potential exposure to indoor allergens in residential environments [1]. Since this seminal publication, an abundant amount of research has been conducted to address these research gaps and some progress has been made to establish consensus methods for sampling indoor allergens [2]. The literature is now replete with studies of allergen sampling devices for air and reservoir sampling, and there are a few excellent literature reviews on personal exposure assessment and sampling related to healthy homes [2,3]. Still, the development of standards for use in assessing the most common household exposures to airborne or surface borne allergens has proved to be elusive. One primary reason for this, as grasped by many researchers who have explored similar domains, such as developing sampling procedures for lead or bacilli spores in dust or air, is understanding the complexities of exposure pathways from the source, to an amplifying reservoir, and to then on to the human target organ.

Why develop a standard on surface sampling for allergens if human exposure pathways from sources to target organs, primarily the respiratory tract, have not been thoroughly established? Should standards on allergen surface sampling be developed, especially if their use in calculating exposure and ultimately risk of allergic sensitization, episodic symptoms of wheezing or other bronchial conditions, development of asthma, or exacerbation of asthma symptoms may not be fully known? This uncertainty notwithstanding, allergy and allergen exposure in the home are likely to have a causal relationship to asthma. Moreover, reservoir dust samples are still used as a surrogate, composite representation of chronic aeroallergen exposure [3]. The epidemiological relationship established between dust mite allergen-exposure and sensitization to this allergen in early life was first established using reservoir sampling [4].

The purpose of this paper is then to provide both a theoretical and evidence based approach toward the ultimate development of a surface sampling standard for indoor aeroallergens in the near term. Clinicians and their patients need

effective, efficient, and evidence based samplers for routine assessment of allergen dust in patients' homes. Health departments and other government agencies may conduct allergen assessments or interventions and need standard means for determining allergen loading or exposures in homes. United States Department of Housing and Urban Development (HUD) developed a vacuum sampling protocol on allergens for its grantees over six years ago [5]. Although the approach provides the concept of convenience and reproducibility, using a vacuum cleaner equipped with an inline filter, its total efficiency for recovery of allergen does not appear to have been scrutinized in the peer review literature [3]. Major studies such as the "The National Children's Study" will examine the effects of environmental influences on the health and development of 100 000 children across the United States, following them from before birth until age 21 [6]. This study, which is expected to launch in the next two years, would benefit from an informed discussion and then standard development for surface borne allergens perhaps using the HUD method with modifications based on more recent findings and discussion [5]. The approach in this paper will include a review of models and empirical studies on allergen exposure assessment and particle dynamics and surface characteristics using a diverse literature on wipe and vacuum sampling inclusive of lead, bacilli spores, endotoxin, and common indoor allergens. Criteria will be identified from theoretical or evidence based considerations for development of allergen-surface sampling standards. Studies that have addressed or failed to meet these criteria during allergen exposure assessment or method development will be identified and the merits of their findings discussed. While a tremendous amount of resources and effort has advanced our molecular and clinical understanding of allergy and asthma, inadequate attention is given to the primary means to assess exposure to indoor allergens through surface sampling and exposure assessment studies. Although the etiology of dust mite and endotoxin induced asthma could be due to a number of reasons, misclassification of exposure could be one of them [7]. The goal of this paper is to inform and encourage the clinical and environmental health-communities to develop allergen-surface sampling standards using a theory and evidence based approach.

Methods

The approach that will be taken here is to (1) describe a rationale for surface sampling of allergens using theories, model based approaches, and best practices; (2) use this rationale to develop criteria for surface sampling; and (3) explore the literature to find evidence base studies that address these criteria. The intent is not to conduct an exhaustive search of all allergen-sampling methods because there are already some excellent reviews that examine airborne as well as surface sampling methods for allergens [2,3]. The intent is also not to evaluate methods for allergen analysis or complete sampling strategies. Excellent resources are available on these subjects [3]. The intent of this paper is to review the literature with a focus both on research gaps and promising developments for establishing performance based standards for the sampling and

collection of surface allergens. Recommendations are provided for advancing the field toward the goal of developing standard, performance-based, allergen surface sampling methods.

Assessment of Indoor Allergens

Arshad, in his recent review of the literature on allergen exposure and the etiology of allergy and asthma, concluded "What we have learned during the past decade is that allergen exposure may cause asthma, be protective, or have no effect, depending on the type of allergen, age of exposure, route of exposure, exposure-dose, and genetic susceptibility" [8]. Does exposure to allergens cause asthma? Answering that question may rely on arriving at improved assessment of aeroallergens whether sampling for airborne or surface allergens. The assessment of aeroallergens is primarily a problem defined by the aerodynamic diameter (AED) of the particle in question. Indoor allergens are carried on household-dust particles and their behavior, at the simplest level, is a matter predominated by the settling velocity of particles. A number of investigations and review articles have developed and summarized our knowledge on the composition of household dust as well as the aerodynamic size of the allergenic particles contained in house dust [2,3,9]. Cat and dog allergens are carried on relatively small aerodynamic particles, 1–10 μm [10–13], dust mite and cockroach allergen are primarily associated with larger particles > 10 μm [10,14,15], fungi can be associated with fungal fragments < 2 μm or spores > 40 μm [16], rodent allergens are on particles > 5 μm [2], and endotoxin, which is common in house dust, is found in both inhalable (< 10 μm) and respirable fractions of air (< 5 μm) [17]. Relatively large particles, such as those containing dust mite or cockroach allergen, settle out of the air quickly, making air sampling difficult but still possible and quantifiable [2,18]. Investigators have considered surface samples of house dust as a composite of potential airborne allergen exposure that in some proportion reach the airways, though in amounts difficult to quantify [2,3]. Some investigators have taken the position that there is little rationale, other than convenience, for collecting surface allergen samples, since allergen collected on a surface is possibly just a surrogate for an airborne exposure to allergen [19]. It is difficult to ignore, however, the past success of surface sampling as a means of exposure assessment for allergen sensitization or symptomatic wheezing [3,7].

There are logical reasons to employ both air and surface sampling for indoor allergens. A long term, low-level exposure to aeroallergens may conceivably take place through chronic exposure to mite allergens in bedding, while a short-term exposure may take place when the bedding is changed and the surface is disturbed, releasing higher levels of allergen. There is evidence for direct inhalation of large particle (radiolabelled pollen, 20 μm diameter) allergen from surfaces, which underscores epidemiological investigations associating dust mite allergen, collected with vacuum dust samples, and sensitization, a process that may average exposures of large particle inhalation over time [20]. Short duration sampling, using air sampling and collection devices, can collect allergen during tasks or activities where dust particles have been resuspended. Air

sampling is also suited for locations where the source of particles are generated over longer periods of time and the particles are of small enough diameter to remain aloft during sampling, such as rodent laboratory facilities, homes with the presence of pets that shed allergen directly into the air, and endotoxin in stables [2,3,17].

Unlike an air sample, which has a straight-forward relationship with an exposure to aerollergens, surface samples, with the exception of direct respiratory contact through bedding, may represent a more complex exposure pathway. Particles that have been generated and released at their source (i.e., dust mite, fungi, endotoxin in a damp and soiled carpet) reside in a reservoir, thus the term "reservoir" dust. Particles that are disturbed or resuspended from this reservoir may settle out of the air thus the term "settled" dust. Unless an investigator is using settling plates or an "A" book sampler, which works like a settled dust collector [21] to sample airborne allergen, than a more precise term on allergen sampling should probably be used to lessen confusion among researchers and practitioners. A more straight-forward term used in this paper is simply "surface dust" or "surface allergen." Either term can represent particulate or allergen which is either or both in the reservoir or settled out of the air. In the absence of a direct mechanism for inhalation a surface sample can represent some composite of a previous airborne exposure. This is not a linear relationship as direct correlations between surface and airborne allergen have been inconsistent [2,22].

Models and Studies that Inform Surface Sampling

Indoor Air Dynamics—Simple and complex models have been developed to explain the relationship between surface, airborne particles and allergens. Most of these models are based on an assumption of conservation of mass that is a quantitative accounting of input and outputs with processes that generate or remove particles inside a hypothetical box or simple room. This one compartment model will mathematically describe a net rate of accumulation of an indoor mass of a particle attribute [23]. This net rate of accumulation is a result of particulate from indoor emissions and particulate supplied from outdoors minus removal from indoor removal processes. The particulate that enters from outdoors follows ventilation pathways that experience losses from mechanical filters and penetration through the room envelope. The indoors will experience particulate loss from deposition and ventilation of particles out of the room. Lai [24], in a review of experimental studies on indoor particle deposition, described 15 chamber or room size experiments on particle removal from deposition. Particle deposition typically follows a pattern with aerodynamic diameters between 0.1 and 1 μm experiencing only slight deposition loss with particles smaller than 0.1 μm depositing at greater frequency due to diffusion as their AED gets smaller. Particles with AEDs greater than 1 μm deposit at greater frequency due to inertial forces as their aerodynamic diameter gets larger. Wallace found deposition of fine and ultrafine particles in an experimental, occupied town house could be reduced by ducted fans and filters [25]. The deposition effects become difficult to model as the complexity of deposition

modifiers, such as electrostatic, surface roughness, turbulent air, and other effects, are known to effect deposition as a function of particle size.

Resuspension—The mirror opposite to particle deposition is resuspension. Carpet and to a lesser extent hard resilient flooring have been considered sinks for house dust. The detachment and re-entry of these particles from these sinks or floors by air currents or a combination of mechanical force or vibration and air currents is called resuspension. Swanson et al. [18] provided a one-compartment model, well known to the clinical community that relates a steady state concentration of allergen to a combined indoor source substance-generation and resuspension rate, which assumes that flooring or other surfaces can both serve as sources as well as sinks for allergen. Thatcher and Layton [26] described a mathematical relationship between the airborne concentration of fine particulate in homes and the process of resuspension. This one-compartment model treats the floor as an independent source of particles activated by resuspension

Resuspension [26]

$$R = \frac{C_{in}(A_d v_d + \lambda_v V) - C_o \lambda_v V}{L_{fl} A_{fl}} \tag{1}$$

where:

R = Resuspension (h^{-1})
C_{in} = Particle concentration indoors (mg m^{-3})
A_d = Interior total deposition surface area (m^2)
v_d = Deposition velocity (m h^{-1})
λ_v = Air exchange rate (h^{-1})
V = Volume (m^3)
C_o = Particle concentration outdoors (mg m^{-3})
L_{fl} = Mass loading of particulate on floor (mg m^{-2})
A_{fl} =floor surface area (m^2)

There are very few studies that have systematically investigated the propensity for particulate matter to be disturbed from hard and textile surfaces. Ferro et al. [27] used a combination of modeling and monitoring of the air to estimate resuspension-source strength from hard and carpeted surfaces using a variety of activities to disturb the dust. They found the majority of airborne particles from these disturbances with AED > 5 μm, however, substantial quantities of dust were generated that were particulate matter less than 2.5 or 5 μm ($PM_{2.5}$ and PM_5). Qian and Ferro followed this work with experiments that considered a two-compartment model, one compartment for airborne pollutant transit, as described previously, and other compartment for resuspending particles entering the air from surfaces and redepositing back to the surface [28]. Using fluorescent, polystyrene plastic-particles as a tracer, a number of activities to disturb the particles were followed by vacuum cleaning. Resuspension was predominately associated with particles greater than 1 μm. Resuspension rate depends on surface loading of pollutants and depth of particles in surfaces.

Qian raised the question of whether only resuspendable particles should be included in estimates of surface loading [28].

The resuspending flux ($\mu g\ m^{-2}\ hr^{-1}$) between the surface dust and deeper dust is complex to model and requires knowledge of the mechanisms that retain and release allergen containing dust from surfaces. Hinds described the primary mechanisms of particle adhesion on ideal surfaces due to van der Waals forces, electrostatic forces, and forces arising from surface tensions of adsorbed liquids on surfaces [29, pp. 141–144]. These forces are affected by particle characteristics, such as composition, shape, surface roughness, and size (diameter) of the particle. The surface will affect adhesion due to its composition, roughness, and contamination of the surface. The microenvironment will in turn affect adhesion through relative humidity, temperature, duration of contact, and initial contact velocity [29, pp. 141–144]. Adhesive forces are proportional to the diameter of the particle, d, while removal forces needed to detach and resuspend the particles are proportional to d^3, such as gravitational, vibrational, and centrifugal forces, and removal forces from air currents are proportional to d^2. As particle size decreases it becomes more difficult to remove particles from surfaces. However, small particles can agglomerate into a thick layer of particles that can easily be dislodged in large pieces [29, pp. 141–144]. When drag forces from air currents and gravitational forces together are greater than adhesive forces, particle can be resuspended in the air. Particles that exceed a critical air velocity can detach and become airborne and this can happen at relatively low velocities for particles that may be moving or sliding [30]. This mechanism illustrates how a vacuum cleaner or a vacuum-powered air sampler, moved across a floor, could resuspend allergen containing particles.

Surface Dynamics—Up to this point, we have described adhesion, detachment, and resuspension of particles without examining the surface structures in a house that will affect the retention and release of allergen and dust. As described by Hinds and later Zhang [30], the characteristics of surfaces including texture and surface roughness will affect the adhesion of particles. Flooring, especially carpet, provide a variety of surface texture that will affect retention and release of particles. Carpet texture or roughness can be evaluated using 3D scanning laser microscopes and deposition of standard particles can correlate with differences in relative area. Altin et al. [31] deposited KCl particles on six carpets which differed in observable properties of pile height and weave and found correlations ($R = 0.9$) with relative areas at extremely fine scales (10^{-4} mm^2) and very coarse scales (10^2 mm).

Carpeting, perhaps the most complex of flooring, since it is observed to be a sink indoors for particles and a reservoir for mites, fungi, and bacteria is difficult to characterize because its retention abilities can change over time with wear [32]. Carpet surface area and fluorocarbon treatment of fibers, however, also have a major affect on the retention and release of dust mite allergens [33].

Wang et al. [34] embedded well characterized leaded dust in a reproducible manner on test carpets and evaluated how a number of parameters, which affect particle adhesion and removal, will also affect the collection of a dust

sample from vacuum sampling. The type of carpet, loop versus shag, relative humidity and resultant electrostatic charge, dust loading, and capture velocity of a vacuum sampler will all affect the collection efficiency of leaded dust.

Roberts [35] assessed surface and deep dust in ten old carpets by vacuuming and using a validated vacuum sampler, HVS4 (ASTM 5438-00, 2000), to sample carpets at various time periods. He assessed deep dust with a vacuum cleaner dirt detector and surface dust with the HVS4. He found that deep dust remains in the carpet after surface dust is removed. He also concluded that removal of surface dust continues to draw deep dust to the surface and often increases the concentration of pollutants by removal of large particles from the surface. This bias for large particle collection during vacuuming [36] could increase concentration of allergen in a carpet after only surface cleaning is performed. If deep dust remaining in a carpet is a major source of surface dust as Roberts suggests should deep dust be assessed as a means to relate chronic exposure to allergen exposure over time? This runs counter to Qian's conclusion, described previously, who considers only surface dust as an airborne risk following activities in the household [28].

The distribution of allergen and dust is exponentially greater from the base to the top of the pile. Causer and Shorter [37] sectioned 24 carpets, primarily wool as found in New Zealand and determined through immunoassay, that dust mite allergen is an order of magnitude greater at the base of the carpet and declines inversely as one reaches the top of the carpet. Although Sercombe [38] found in a separate study of five used carpets, that the cross-sectional distribution of allergen throughout carpets was highly variable, allergen was at least 1 order of magnitude greater at the base of the carpet than at the surface of the carpet.

Metrics for Measuring Surface Allergen—The importance of dust loading in an assessment of allergen exposure is a subject of controversy. Allergen loading is the product of allergen concentration (μg/g) and dust mass per area (g/m^2). Some investigators take the position that dust mass correlates well with allergen content and take the position there is no need to determine mass of dust per area sampled. Most major national studies use concentration only in expressing exposure. Arbes [39], in a study to determine the feasibility of study participants taking their own dust samples, determined that participants usually biased the collection toward collecting a smaller mass of dust than technicians using standard methods. Lewis [36] took the position that some vacuum samplers may have a large particle bias and do not sample-small allergen containing particles as efficiently as large particles. "A consequence of using a typical handheld vacuum sampler, followed by data reporting of allergen mass per mass of bulk dust collected, may be an underestimation of exposure due to a relative underestimation of the numerator (allergen recovery) with respect to the denominator (dust recovery)." Unless someone was to sample fine-homogenous dust, say from a mattress, where the allergen collected and the dust collected are close in particle diameter (as opposed to a carpet), dust loading, which measures area in the denominator, should be assessed [36]. Other studies,

however, indicate that allergen concentration is still more correlated to airborne concentrations than allergen loading [21,22]. The improved and standardized methods of surface and air sampling for allergens may further illuminate the best metric for reservoir allergen and clinically relevant exposure [22].

Lioy [40] in a review of using dust as an exposure metric (149 references) states that "the major challenge for exposure analysis is ensuring that the sampler retrieves material that is indicative of the types and levels that a person comes into contact with on the rug." This also includes material that can be dislodged and resuspended. Lioy further states that "The efforts to establish performance evaluations and determine what a dust sample represents must become part of the process for development and selection of samples for use in research, regulating, and forensic investigations" [40].

Criteria for Developing Performance Based Standard for Sampling and Collection of Surface Allergens—Lioy stated for detailed exposure assessments a very well characterized vacuum sampler should account for particle-size fractionated mass distributions [40]. As described in previous sections of this paper, dust and allergen containing dust will distribute in specific ways within a carpet and be retained on surfaces due to a number of adhesive mechanisms. Whether a vacuum sampler or a wipe sampler is used to collect allergen from surfaces, specific performance criteria should be set that is informed by criteria critical to ensure accurate, precise, and reproducible measurements.

Performance based test methods define general approaches for sampling, sample preparation, and making measurements on a specified material. They set maximum allowable uncertainties for each component of uncertainty of each measured constituent over its validated concentration range. The key criteria of compliance with a performance-based standard test method is the quality of data generated rather than adherence to procedure" [41]. Performance criteria for air sampling has also been described [42]. Criteria for surface sampling should include some primary criteria for air sampling [43]. Criteria should be based on both mechanistic considerations, accuracy, precision, bias, reproducibility, ease of use, and cost. The later two categories are important since use of a heavy and expensive surface sampler will reduce the ability and willingness of study participants or health departments from conducting extensive sampling.

Theory or Mechanism Based—An allergen surface sampler should have a rationale for its use as described by Lioy [40]. An allergen-surface sample should represent a risk based on a portion of it that comes into contact with a person. This criterion should then be based on whether a chronic or acute exposure to allergens is assessed. Hawkins [44], in describing sample-averaging times for occupational exposure assessment, said when brief exposure causes health effect, shorter sample times are meaningful. When a substance accumulates in the body with little or no removal or transformation, a long term time weighted average exposure over months or years may be appropriate [44]. Although this guideline was intended for occupational exposures, it clearly parallels the issue of acute and chronic health effects from allergens. For example, episodic

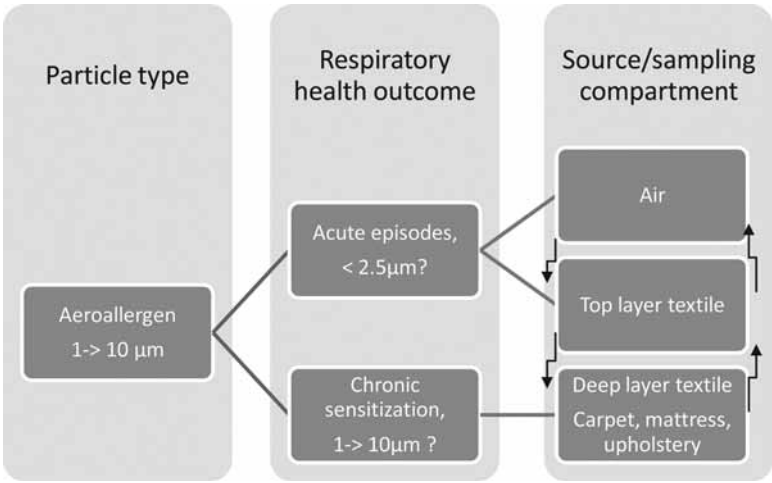

FIG. 1—*Simple hypothesis for allergen surface sampling based on exposure pathways and risk (angular connecting lines between compartments indicate partial linear relationship).*

wheezing could be due to a brief, resuspended allergen exposure from the surface of a carpet. The same type of acute episode, however, could come about from an exposure to allergen from the deep dust within a mattress or carpet. The transfer of allergen through a textile, especially after incomplete cleaning, could result in this type of exposure. A chronic health effect, such as sensitization to an allergen could conceivably happen over a period of time and a surface sample from throughout a surface, such as a carpet or mattress, might best be represented here. This is the deep dust as described by Roberts et al. [35] that could be brought to the surface over time. Contact with the surface might be a direct form of exposure to the head region of the upper airways. This could conceivably occur from sleeping in bedding where there are large exposures to dust mite allergen [20]. This very simple risk based to surface sampling of allergen is illustrated in Fig. 1. Surface sampling may also be used to assess environmental interventions and in this case a sampler that can evaluate superficial from deep cleaning of surfaces would be valuable. This risk based approach, however, is still only a hypothesis. The complexity of the relationship between the three compartments in Fig. 1, as described well by Tovey et al., is linear in some circumstances [21]. The non-linearity between compartments is also supported by evidence [22] and Tovey et al. concludes that the difference between his findings of linearity in these compartments and other studies that indicate non-linearity may be explained due to statistical power in sampling high concentration dust mite allergen, such as can be found in Sydney, Australia, longer sampling times for all types of allergen, and degree of disturbance of the reservoir. The "angular lines" drawn between the compartments in Fig. 1 are meant to illustrate the uncertain association between the compartments.

Accuracy—Accuracy can be defined "as the degree of correctness with which a measurement system yields the true value of an observable" [42, pp. 201–231]. Specifically, this is the percent difference between the measured and true values. Accuracy for a surface sampler will be defined here as some degree of correctness expressed by the total efficiency. A total efficiency will be defined as the product of the collection and sampling efficiency. Either the sampler or collector could be altered to achieve a high total efficiency.

Sampling efficiency—the ratio of a mass of particles (or particle number) removed by a sampler to mass (or particle number) of particles deposited or embedded in surface.

Collection efficiency—the ratio of the mass of particles (or particle number) collected in a filter, cyclone, or other collection media to the mass removed from the sampler.

The total, collection, or sampling efficiency is typically expressed as a percent.

• Efficiency of a hypothetical allergen surface sampler

$$FC/E = (FC/R)(R/E) \tag{2}$$

Total efficiency = FC/E,
Collection efficiency = (FC/R),
Sampling efficiency = (R/E),
FC = mass retained in filter or cyclone other media,
E = mass deposited or embedded in surface, and
R = mass removed from surface

Sampler Performance Criteria—The sampler should have sufficient capture velocity, if a vacuum sampler, or sufficient adhesive qualities, if a wipe sampler, to remove a given amount of mass or particles from a surface. Examples of the wipe sampler for allergen might be the electrostatic cloth sampler or the press tape sampler [22]. A range of artificial, reproducible, surface materials with representative textures of household material should be developed in order to standardize sampling efficiency between sampling devices [40,45].

Collection Performance Criteria—Filters or cyclones or other media should have the ability to retain sample removed by a collector. Filters and cyclones and other media are tested and validated for particle collection efficiency and manufacturer's information should be evaluated [42,46, pp. 281–314].

Total Efficiency Performance Criteria—The total efficiency should be evaluated by particle mass at each diameter, i.e., particle-size mass. The Baltimore Repair and Maintenance sampler had a total collection efficiency of 87% for talcum dust, in the size range of 0.5–44 μm [47]. This may be considered a starting point for discussion of total collection efficiency for allergen particles.

Precision—The standard deviation of repeated measurements of the same observable with the same measurement method [42, pp. 201–231]. Also, a

relative standard deviation (coefficient of variation) could be computed for an estimate of precision.

Precision Performance Criteria—To optimize precision, the sampler should have protocols or device aids to estimate pressure (such as gauges to estimate static or velocity pressure entering sampler) for use on an array of surfaces. Some measure of surface roughness should be recorded for each surface sampled. A qualitative measure could be used in the absence of a practical means to ensure quantitative information on surface roughness or retention characteristics of surfaces that are sampled.

Bias—Ratio of measured value to the true value is reflected primarily in the sampler efficiency but also in collection efficiency [42, pp. 201–231]. A bias can occur where the sampler or collector under or over performs on a specific particle-size mass fraction. For example, if the sampler nozzle is not correctly configured, it might undersample a specific mass of a particle size distribution, such as mite fecal particles, 10–40 μm in diameter. The bias may also be an objective to achieve, if desired. For example, it may be desired to bias particles toward collection of those at the surface or particles of a specific size, say in the respirable range, similar to size-selective sampling in air sampling criteria [42, pp. 217–229].

Bias Performance Criteria—An air sampler can experience bias from gravitational and inertial forces. The bias from a vacuum sampler is assumed to be largely from inertial mechanisms [29, pp. 213–214]. The bias or particle entering a sampling probe or sampler nozzle increases with increasing particle size and higher inlet velocities. To minimize bias, a sampler inlet can be designed for a specific volumetric flowrate, and aerodynamic diameter, with a minimal probe opening (diameter in millimeters). This makes use of Davies criteria [29, pp. 213–214]. Particle bias by size-mass fraction could also occur due to transport losses of particles from the surface to the final collection of allergen. Reducing transport paths, reducing bends in these paths, and keeping these transport paths as short as possible are desired criteria [29, pp. 216–217].

Reproducibility—The extent to which a method yields the same response to the same quantity of pollutant [46].

Reproducibility for allergen surface sampling might be the manner in which sampling is generally unaffected or has minimal impact from electrostatic charge of the surface. For example, Wang [34] pointed to relative humidity that caused differential results with a vacuum sampler for lead dust. Ensuring that a sampler would not collect an electrostatic charge during sampling could be important for ensuring a reproducible outcome. Also, ensuring that filters are not unduly influenced by overloading and produce only negligible resistance to flow during routine use is expected to increase reproducibility. The need to change the filter during the sampling process could introduce error and reduce sampling reproducibility.

Ease of Use—The ability to use allergen surface samplers by study participants, homeowners, and modestly trained-technicians is an important criteria [39]. Some research studies may require a higher level of accuracy or even precision than what is needed for a clinical screening or semi-quantitative use of allergen surface samplers. The variation in the estimate of exposure may be best served by taking more measurements with less inherent accuracy and precision than fewer measurements with more accuracy and precision.

Cost—The cost of some surface samplers may exceed $2000, such as an HVS3 or HVS4. The cost of a thimble with filter is inexpensive and the DUSTREAM sampler and filter costs only $12. Ensuring a reasonable cost is an important criterion. However, it may not be possible for an inexpensive sampler to match the collection ability of more expensive samplers.

Literature Search on Published Documents that Demonstrate Surface Sampler Criteria—A literature search of surface sampling methods, inclusive of lead, bacillus, endotoxin, and allergens was performed to evaluate laboratory or field based studies that evaluated these criteria based on an evidence based approach used in public health. Databases such as PUBMED were used and key words such as allergen, sampler, vacuum sampling, dust sampling were put in search engines. Documents available from government websites, such as HUD's documents were obtained from the HUD Office of Healthy Home's website in Washington, D. C., http://www.hud.gov/offices/lead/. The evidence base approach relies on a statement of the issue, determination of what is known in the scientific literature, and a quantification of the issue [48]. Wherever possible, peer reviewed papers were sought that tested surface samplers and used quantifiable approaches to evaluate the criteria previously described. Similar to an evidence based approach to public health, an action plan will be recommended to take the performance criteria to the scientific community for comment and feedback.

Table 1 below lists papers that were found that met certain criteria as described for development of a performance standard for allergen surface sampling. A few papers are listed that provide useful comparative information on sampling for lead and other metals. Many papers were found that described sampling device correlations with other methods or with blood serum IgE [21,22,49]. These papers are not listed in Table 1 since they fail to show defined accuracy and precision. A few national studies are presented in Table 1, which although failing the criteria, are of significant importance because of their national or international reach.

Recommendations for Development of Performance Based Standard—Development of an ASTM method that takes into account the sampling criteria, if not all, but most described in the previous section, would help establish a mechanism driven, accurate, and precise allergen surface sampling method that could be used in national or international studies. Many of the national and international studies on allergy and asthma could be improved by

TABLE 1—*Studies that have fulfilled various criteria for allergen surface sampling.*

Author and reference	Sampler/ Collector	Mechanism	Accuracy	Precision	Bias	Reproducible	Ease of use	Cost
Sercombe [22]	Electrostatic cloth	Surface adhesion	1.44 vs. 4.24 $\mu g/m^2$ against reference vacuum	Range $\mu g/m^2$ 1.44± (0.38–19.46)	Spearman r_s (*P*-value) with reference vacuuming method, 0.62 (0.0103)		Easy	< $1.00
Sercombe [22]	Press-tape	Surface adhesion	13.92 vs. 4.24 $\mu g/m^2$ against reference vacuum	Range $\mu g/m^2$ 1.44± (2.08–81.50)	Spearman r_s (*P*-value) with reference vacuuming method, 0.55(0.0283)		Easy	<$1.00
Farfel, M [47]	Plastic nozzle, steel cyclone	Lead, surface layer	87% 0.5–44 μm, talc, 89% 38–149 μm	—	Cyclone, >97% housedust 5μm cutpoint	—	—	—
Lewis, RD [36]	Plastic Handheld vacuum, bonnet filter	Dust Mite Allergen from embedded carpets, surface layer	Unkown, allergen was 4× greater with HVS3 than Readivac	Readivac 6735, 27% HVS3, 21%	*r* for bulk dust and allergen=0.54; *r* for bulk dust and allergen 0.91	Readivac, plastic housing and transport loss	Handheld	< $50
Upright vacuum, steel nozzle, cyclone	HVS3, polished anodized interior	Bulky	<$3,000					

TABLE 1—*Continued*

Author and reference	Sampler/ Collector	Mechanism	Accuracy	Precision	Bias	Reproducible	Ease of use	Cost
Wang, [34]	Plastic pvc pipe nozzle, vacuum motor, filter bag polyethylene and polypropylene	Lead, Surface layer	83% recovery, level loop, 60% RH, 8g/m^2 loading	83 ± 4 %	—	Evaluated effects of relative humidity, loading, electrostatic charge, vacuum velocity Used in national lead study (CLEARS)	Somewhat bulky	unknown
Vojta National Survey of Lead and allergens in Housing [50]	Mitey Mite canister vacuum/ whatman filter thimble	Surface layer	—	—	—	Validated in national study	Wand, easy	< $100?
ISAAC 2004 International Study of Asthma and Allergies in Childhood [51]	Vacuum cleaner 800 w motor, plastic wand, ALK filter	surface	Filter retains 74% particles, 0.3-0.5 μm, 81% of particles 0.5-1.0 μm, 100% > 1 μm	—	—	Validated in national study	Easy to use	<$100?

TABLE 1—*Continued*

Author and reference	Sampler/ Collector	Mechanism	Accuracy	Precision	Bias	Reproducible	Ease of use	Cost
Ashley, K [52]	Portable sampling pump, ASTM D7144, 37mm filter cassette, 0.8 μm pore PVC	Metals, powdered leads based paint	59.2%, glass surface, three loadings,	59.2 ±11.0%, 19% CV	—	Validated standard	Portable	Cost of pump, $500
Surface	Other rough surfaces measured							

Note: Costs of some samplers were estimates based on design of sampler; Some data not available = —; Accuracy percentages are % recovery or efficiency; Precision is either relative standard deviation (coefficient of variation) or mean ± std. or range; Bias, r = correlation coefficient.

establishing criteria, which does not significantly alter the inexpensive and easy to use sampling devices. Future research and development should be directed toward making sampling nozzles that minimize electrostatic charge, use of appropriate inlet size and volumetric flow to reduce particle bias, and total collection efficiency that should meet 80% or higher recovery of allergen-particles in the respirable range, similar to what Farfel established, on smooth surfaces with the cyclone studies in 1994 [47] or what Wang [34] determined from level loop carpets. Use of standard surfaces and standard dusts are needed to further develop samplers. Samplers also need the ability to adjust to these different surfaces and gauges that can be situated to ascertain applied pressure, such as static pressure at the nozzle, would be ideal. Finally, serious consideration should be given to conducting more studies where simple but potentially useful press tape and electrostatic samplers can be used for assessing surface allergen available for rapid release in the residential environment [22].

Conclusion

One of the findings from this review is that criteria for allergen-surface sampling, based on theoretical considerations and evidence based studies, have not been widely adopted in studies relating exposure to allergies or asthma. Recommendations have been made by a number of authors [2], since the IOM publication [1], to standardize surface sampling of allergens but only a small amount of progress to date has been made toward this goal. Consensus around a few samplers, notably the Eureka vacuum canister equipped with inline filters, either Whatman or proprietary such as the DUSTREAM, have been developed but insufficient work has been done to characterize these by the criteria advanced in this paper. One barrier toward establishment of standardization may be due to a lack of agreement on the underlying scientific basis for allergen surface sampling. A theoretical and evidence based foundation for development of performance base standards has been offered in this paper. Criteria for performance sampling have been suggested. Sampling collection devices widely used today for allergen surface sampling may be acceptable and may meet or exceed the criteria as discussed in this paper. It may prove valuable to evaluate all allergen surface samplers around accepted criteria of accuracy and precision, and doing so may help to elucidate the relationship between allergens and clinical disease.

Acknowledgments

The writers would like to acknowledge the efforts of Kenyetta Daniels and Xue Liu, graduate research assistants at the Environmental Health Research Laboratory, Saint Louis University School of Public Health, for assistance in the literature search and development of the EndNote library for this manuscript. We would also like to thank the San Destin Consulting Group (J.L., R.S., S.S., M.S., and L.S.) for timely advice.

References

[1] *Indoor Allergens: Assessing and Controlling Adverse Health Effects*, Pope, A. M., Patterson, R., and Burge, H., Eds., Committee on the Health Effects of Indoor Allergens, Institute of Medicine, National Academy Press, 1993, Washington, D.C.

[2] O'Meara, T. and Tovey, E., "Monitoring Personal Allergen Exposure," *Clin. Rev. Allergy Immunol.*, Vol. 18, No. 3, 2000, pp. 341–395.

[3] Peter Ashley, J. R. M., Maureen, A. W., Joanna, G., and Jack, A., *Healthy Home Issues: Residential Assessment*, HUD Office of Healthy Homes and Lead Hazard Control, 2006.

[4] Sporik, R., Holgate, S. T., Platts-Mills, T. A., and Cogswell, J. J., "Exposure to House-Dust Mite Allergen (Der p I) and the Development of Asthma in Childhood. A Prospective Study," *N. Engl. J. Med.*, Vol. 323, No. 8, 1990, pp. 502–507.

[5] Battelle, *Vacuum Dust Sample Collection Protocol for Allergens*, Q. Tech, Ed., HUD Office of Healthy Homes and Lead Hazard Control, Washington, D.C., 2008.

[6] Landrigan, P. J., Trasande, L., Thorpe, L. E., Gwynn, C., Lioy, P. J., D'Alton, M. E., Lipkind, H. S., Swanson, J., Wadhwa, P. D., Clark, E. B., Rauh, V. A., Perera, F. P., and Susser, E., "The National Children's Study: a 21-year prospective study of 100,000 American children," *Pediatrics*, Vol. 118, No. 5, 2006, pp. 2173–2186.

[7] Celedon, J. C., Milton, D. K., Ramsey, C. D., Litonjua, A. A., Ryan, L., Platts-Mills, T. A., and Gold, D. R., "Exposure to Dust Mite Allergen and Endotoxin in Early Life and Asthma and Atopy in Childhood," *J. Allergy Clin. Immunol.*, Vol. 120, No. 1, 2007, pp. 144–149.

[8] Arshad, S. H., "Does exposure to indoor allergens contribute to the development of asthma and allergy?," *Curr. Allergy Asthma Rep.*, Vol. 10, No. 1, 2010, pp. 49–55.

[9] Luedtke, A. E., Floor Covering, *Dust and Airborne Contaminants*, 2003.

[10] de Blay, F., Heymann, P. W., Chapman, M. D., and Platts-Mills, T. A., "Airborne Dust Mite Allergens: Comparison of Group II Allergens With Group I Mite Allergen and Cat-Allergen Fel d I," *J. Allergy Clin. Immunol.*, Vol. 88, No. 6, 1991, pp. 919–926.

[11] Custovic, A., *et al.*, "Distribution, Aerodynamic Characteristics, and Removal of the Major Cat Allergen Fel d 1 in British Homes," *Thorax*, Vol. 53, No. 1, 1998, pp. 33–38.

[12] Custovic, A., Green, R., Fletcher, A., Smith, A., Pickering, C. A., Chapman, M. D., and Woodcock, A., "Aerodynamic Properties of the Major Dog Allergen Can f 1: Distribution in Homes, Concentration, and Particle Size of Allergen in the Air," *Am. J. Respir. Crit. Care Med.*, Vol. 155, No. 1, 1997, pp. 94–98.

[13] Luczynska, C. M., Li, Y., Chapman, M. D., and Platts-Mills, T. A. E., "Airborne Concentrations and Particle Size Distribution of Allergen Derived From Domestic Cats (Felis domesticus). Measurements Using Cascade Impactor, Liquid Impinger, and a Two-site Monoclonal Antibody Assay for Fel d I," *Am. Rev. Respir. Dis.*, Vol. 141, No. 2 I, 1990, pp. 361–367.

[14] Pope, A. M., "Indoor Allergens—Assessing and Controlling Adverse Health Effects," *JAMA*, Vol. 269, No. 21, 1993, p. 2721.

[15] De Lucca, S. D., Taylor, D. J., O'Meara, T. J., Jones, A. S., and Tovey, E. R., "Measurement and Characterization of Cockroach Allergens Detected During Normal Domestic Activity," *J. Allergy Clin. Immunol.*, Vol. 104, No. 3 Pt 1, 1999, pp. 672–680.

[16] Burge, H. A., "Aerobiology of the Indoor Environment," *Occup. Med.*, Vol. 10, No. 1, 1995, pp. 27–40.

[17] Von Mutius, E., Braun-Fahrlander, C., Schierl, R., Riedler, J., Ehlermann, S., Maisch, S., Waser, M., and Nowak, D., "Exposure to Endotoxin or Other Bacterial

Components Might Protect Against the Development of Atopy," *Clin. Exp. Allergy*, Vol. 30, No. 9, 2000, pp. 1230–1234.

[18] Swanson, M. C., Campbell, A. R., Klauck, M. J., and Reed, C. E., "Correlations Between Levels of Mite and Cat Allergens in Settled and Airborne Dust," *J. Allergy Clin. Immunol.*, Vol. 83, No. 4, 1989, pp. 776–783.

[19] Renstrom, A., "Exposure to Airborne Allergens: A Review of Sampling Methods," *J. Environ. Monit.*, Vol. 4, No. 5, 2002, pp. 619–622.

[20] Lewis, R. D., Sterling, D. A., King, B., Hendershott, L., Goodgold, H., and Hutcheson, P., "Evidence of large particle allergen inhalation from contact with surfaces," *Ann. Allergy Asthma Immunol.*, Vol. 83, No. 1, 1999, pp. 41–48.

[21] Tovey, E. R., Mitakakis, T. Z., Sercombe, J. K., Vanlaar, C. H., and Marks, G. B., "Four Methods of Sampling for Dust Mite Allergen: Differences in 'Dust'," *Allergy*, Vol. 58, No. 8, 2003, pp. 790–794.

[22] Sercombe, J. K., Liu-Brennan, D., Garcia, M. L., and Tovey, E. R., "Evaluation of Home Allergen Sampling Devices," *Allergy*, Vol. 60, No. 4, 2005, pp. 515–520.

[23] Nazaroff, W. W., "Indoor Particle Dynamics," *Indoor Air*, Vol. 14, No. Suppl 7, 2004, pp. 175–183.

[24] Lai, A. C. K., "Particle Deposition Indoor: A Review," *Indoor Air*, Vol. 12, No. 4, 2002, p. 211–214.

[25] Wallace, L. A., Emmerich, S. J., and Howard-Reed, C., "Effect of Central Fans and In-Duct Filters on Deposition Rates of Ultrafine and Fine Particles in an Occupied Townhouse," *Atmos. Environ.*, Vol. 38, No. 3, 2004, pp. 405–413.

[26] Thatcher, T. and Layton, D., "Deposition, Resuspension, and Penetration of Particles Within a Residence," *Atmos. Environ.*, Vol. 29, No. 13, 1995, pp. 1487–1497.

[27] Ferro, A. R., Kopperud, R. J., and Hildemann, L. M., "Source Strengths for Indoor Human Activities that Resuspend Particulate Matter," *Environ. Sci. Technol.*, Vol. 38, No. 6, 2004, pp. 1759–1764.

[28] Qian, J., Ferro, A. R., and Fowler, K. R., "Estimating the Resuspension Rate and Residence Time of Indoor Particles," *J. Air Waste Manage. Assoc.*, Vol. 58, No. 4, 2008, pp. 502–516.

[29] Hinds, W. C., *Aerosol Technology Properties, Behavior, and Measurement of Airborne Particles*, 2nd ed., Wiley-InterScience Publication, New York, 1999, p. 483.

[30] Zhang, Y., *Indoor Air Quality Engineering*, CRC, Boca Raton, FL, 2005, p. 615.

[31] Altin, C., Huynh, T., Powers, B., Brown, C. A.,Wang, S., and Zhang, J. S., "Assessing surface roughness for correlation with particulate deposition in IAQVEC 2010," *The Seventh International Conference on Indoor Air Quality, Ventilation and Energy Conservation in Buildings*, Syracuse, N.Y, 2010.

[32] Causer, S. M., Lewis, R. D., Batek, J. M., Sr., and Ong, K. H., "Influence of Wear, Pile Height, and Cleaning Method on Removal of Mite Allergen From Carpet," *J. Occup. Environ. Hyg.*, Vol. 1, No. 4, 2004, pp. 237–242.

[33] Lewis, R.D., Breysse, P. N., Lees, P. S., Diener-West, M., Hamilton, R. G., andEggleston, P., "Factors Affecting the Retention of Dust Mite Allergen on Carpet," *Am. Ind. Hyg. Assoc. J.*, Vol. 59, No. 9, 1998, pp. 606–613.

[34] Wang, E., Rhoads, G. G., Wainman, T., and Lioy, P. J., "Effects of Environmental and Carpet Variables on Vacuum Sampler Collection Efficiency," *Appl. Occup. Environ. Hyg.*, Vol. 10, No. 2, 1995, pp. 111–119.

[35] Roberts, J. W., Glass, G., and Mickelson, L., "A Pilot Study of the Measurement and Control of Deep Dust, Surface Dust, and Lead in 10 Old Carpets Using the 3-Spot Test While Vacuuming," *Arch. Environ. Contam. Toxicol.*, Vol. 48, No. 1, 2004, pp. 16–23.

[36] Lewis, R. D. and Breysse, P. N., "A Comparison of the Sampling Characteristics of Two Vacuum Surface Samplers for the Collection of Dust Mite Allergen," *Appl. Occup. Environ. Hyg.*, Vol. 13, No. 7, 1998, pp. 536–541.
[37] Causer, S., Shorter, C., and Sercombe, J., "Effect of floorcovering construction on content and vertical distribution of house dust mite allergen, Der p I," *J. Occup. Environ. Hyg.*, Vol. 3, No. 4, 2006, pp. 161–168; quiz D45.
[38] Sercombe, J. K., Liu-Brennan, D., Causer, S. M., and Tovey, E. R., "The Vertical Distribution of House Dust Mite Allergen in Carpet and the Effect of Dry Vacuum Cleaning," *Int. J. Hyg. Environ. Health*, Vol. 210, No. 1, 2007, pp. 43–50.
[39] Arbes, S. J., Sever, M., Vaughn, B., Mehta, J., Lynch, J. T., Mitchell, H., Hoppin, J. A., Spencer, H. L., Sandler, D. P., and Zeldin, D. C., "Feasibility of Using Subject-Collected Dust Samples in Epidemiologic and Clinical Studies of Indoor Allergens," *Environ. Health Perspect.*, Vol. 113, No. 6, 2005, pp. 665–669.
[40] Lioy, P. J., Freeman, N. C. G.,and Millette, J. R., "Dust: A Metric for Use in Residential and Building Exposure Assessment and Source Characterization," *Environ. Health Perspect.*, Vol. 110, No. 10, 2002, pp. 969–983.
[41] ASTM. Committee E01 Preparing to Implement Performance-Based Methods, 2004 www.astm.org/SNEWS/AUGUST_2004/methods_aug04.html.
[42] *American Conference of Governmental Industrial Hygienists, Air Sampling Instruments for Evaluation of Atmospheric Contaminants*, 9th ed., ACGIH Worldwide. x, Cincinnati, Ohio, 2001, p. 740.
[43] *NIOSH, NIOSH Manual of Analytical Methods*, NIOSH, 2003.
[44] Neil C Hawkins, S. K. N. and James, C. R., *A Strategy for Occupational Exposure Assessment*, American Industrial Hygiene Association, Fairfax, VA, 1991, p. 46.
[45] Edmonds, J. M., "Efficient Methods for Large-Area Surface Sampling of Sites Contaminated With Pathogenic Microorganisms and Other Hazardous Agents: Current State, Needs, and Perspectives," *Appl. Microbiol. Biotechnol.*, Vol. 84, No. 5, 2009, pp. 811–816.
[46] *American Conference of Governmental Industrial Hygienists, Air Sampling Instruments for Evaluation of Atmospheric Contaminants*, 6th ed., ACGIH Worldwide, Cincinnati, Ohio, 1989.
[47] Farfel, M. R., Lees, P. S. J., Rohde, C. A., Lim, B. S., Bannon, D., and Chisolm, Jr., J. J., "Comparison of a Wipe and a Vacuum Collection Method for the Determination of Lead in Residential Dusts," *Environ. Res.*, Vol. 65, No. 2, 1994, pp. 291–301.
[48] Brownson, R. C., Gurney, J. G.,and Land, G. H., "Evidence-Based Decision Making in Public Health," *J. Public Health Manage. Pract.*, Vol. 5, No. 5, 1999, pp. 86–97.
[49] Wickens, K., Lane, J., Siebers, R., Ingham, T., and Crane, J., "Comparison of Two Dust Collection Methods for Reservoir Indoor Allergens and Endotoxin on Carpets and Mattresses," *Indoor Air*, Vol. 14, No. 3, 2004, pp. 217–222.
[50] Vojta, P. J., Randels, S. P., Stout, J., Muilenberg, M., Burge, H. A., Lynn, H., Mitchell, H., O'Connor, G. T., and Zeldin, D. C., "Effects of Physical Interventions on House Dust Mite Allergen Levels in Carpet, Bed, and Upholstery Dust in Low-income, Urban Homes," *Environ. Health Perspect.*, Vol. 109, No. 8, 2001, pp. 815–819.
[51] Module 4.1—Sampling of Dust for Determination of Allergen Content, 2004, http://isaac.auckland.ac.nz./phases/phasetwo/phasetwomodules.pdf.
[52] Ashley, K., *et al.*, "Evaluation of a Standardized Micro-Vacuum Sampling Method for Collection of Surface Dust," *J. Occup. Environ. Hyg.*, Vol. 4, No. 3, 2007, pp. 215–223.

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Reprinted from JAI, Vol. 8, No. 4
doi:10.1520/JAI103390
Available online at www.astm.org/JAI

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Evaluation of a Handwipe Disclosing Method for Lead

ABSTRACT: A qualitative chemical screening method for lead in wipe samples was evaluated for its utility in detecting the presence of lead in collected dust; preliminary evaluation of the performance of the method is reported here. In evaluating the method on pure lead compounds, the observed intensity of the characteristic color change due to the presence of lead was generally consistent with the relative solubilities of the tested compounds. Some pure (non-lead) metal compounds (e.g., those of Ag, Ba, Bi, Ca, Cd, Hg, and Sr) were found to give false positive results. Several representative lead-containing reference materials were also tested, and the qualitative test results differed for different materials. For materials collected on wipes, the method was found to be effective for detecting lead in several sample matrices commonly found in occupational settings. The technique was also applied on-site on dermal samples collected at field locations.

KEYWORDS: handwipe, lead, qualitative analysis, screening, workplace

Manuscript received September 27, 2010; accepted for publication March 1, 2011; published online April 2011.

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Presented at the ASTM International Symposium on Surface and Dermal Sampling, Oct. 14–15, 2010, San Antonio, TX.

Cite as: Ashley, K., Wise, T. J. and Esswein, E. J., "Evaluation of a Handwipe Disclosing Method for Lead," *J. ASTM Intl.*, Vol. 8, No. 4. doi:10.1520/JAI103390.

Introduction

Qualitative field screening methods are sometimes used in environmental health and occupational hygiene applications for the on-site detection of toxic substances of concern, e.g., heavy metals such as lead [1–3]. Such methods may be used to assess whether toxic agents of concern are present or absent, even with respect to regulatory action levels [4]. Field screening tests can provide a timely means for assessing potential human exposures to toxic materials, but their validity is called into question if their performance has not been evaluated or verified [5,6].

A qualitative colorimetric (screening) method for disclosing the presence of lead in dust wipe samples has been developed [7,8]. The method entails the use of a wetted handwipe to collect dust on surfaces such as skin, floors, and myriad other substrates that are potentially contaminated with lead. The wipe is subsequently treated with a weakly acidic leaching solution in order to extract soluble lead that may be present in the collected dust matrix. The presence of lead is then disclosed by use of an aqueous solution of rhodizonic acid, which forms a characteristic pink- to red-colored complex under acidic conditions [9,10].

The goal of this study was to evaluate the handwipe disclosing technique for lead by subjecting the method to a series of validation tests. In previous work, we have evaluated qualitative colorimetric screening methods for lead in air filter samples [11] and in paint coatings [12]. Because the performance characteristics of qualitative spot tests are matrix-dependent [13,14], it is recommended to evaluate these techniques for each analytical matrix of interest. In the present study, we have evaluated the response of the handwipe screening method for lead using chemically pure standards of known lead compounds. Also, the method was tested on representative matrix chemical standards, i.e., certified reference materials (CRMs), and on-site on dermal samples collected in field locations where lead was a suspected contaminant.

Experimental

Reagents

All chemicals used in this work were reagent grade. Rhodizonic acid (disodium salt), $Ca(NO_3)_2$, KCl, K_2CrO_4, KNO_3, NaCl, Pb_3O_4, PbS, TlCl, and $TlNO_3$ were obtained from Sigma-Aldrich (Milwaukee, WI). Al_2O_3, $BaCO_3$, $Cd(NO_3)_2$, and $NaNO_3$ were from J. T. Baker (Philipsburg, NJ). PbO and $PbBr_2$ came from Johnson Matthey (Royston, Herts., United Kingdom). Pb metal (granular) and PbO_2 were from Merck (Rahway, NJ). $AgNO_3$, $Al(NO_3)_3$, Fe_2O_3, $Hg(NO_3)_2$, K_2SO_4, $PbCrO_4$, and $PbSO_4$ were purchased from Fisher Scientific (Fair Lawn, NJ). AgCl, $PbCl_2$, $Pb(NO_3)_2$, Pb_3O_4, and $Sr(NO_3)_2$ were obtained from Mallinckrodt (St. Louis, MO). $Cd(CH_3COO)_2$, $Pb(CH_3COO)_2$, and Te powder came from Matheson, Coleman, and Bell (Norwood, OH). $Ba(NO_3)_2$, $BaCl_2$, $Bi(NO_3)_3$, $Ca(CH_3COO)_2$, $Mg(NO_3)_2$, $NiCl_2$, $Ni(CH_3COO)_2$, and $Zn(CH_3COO)_2$ were from

Chem Service (Media, PA). Metal standard solutions (≈1000 μg/mL) were obtained from Inorganic Ventures (Lakewood, NJ).

Deionized water (18 MΩ) was produced using a Barnstead Nanopure system (Thermolyne, Dubuque, IA). White vinegar (5 % aqueous acetic acid) was purchased from a local grocery store (Bigg's, Harrison, OH).

Materials

Dust wipes meeting standard performance specifications [15] were obtained from Palintest (Lot No. 080245; Gateshead, Tyne, and Wear, United Kingdom). Wax paper was found in a local convenience store (Ameristop, Barnesburg, OH). Disposable nitrile laboratory gloves were ordered from Fisher Scientific. Plastic pump spray bottles (vol. ≈150 mL) yielding fine-mist aerosols were purchased from U.S. Plastics (Lima, OH).

Representative lead-containing CRMs consisted of National Institute of Standards and Technology (NIST) Standard Reference Materials® (SRMs) (NIST, Gaithersburg, MD). The SRMs used were as follows: (a) 1579a, lead-based paint; (b) 1633b, coal fly ash; (c) 1648, urban particulate matter; (d) 2580, powdered paint I; (e) 2581, powdered paint II; (f) 2582, powdered paint III; (g) 2583, trace elements in indoor dust I; (h) 2584, trace elements in indoor dust II; (i) 2709, San Joaquin soil; (j) 2710, Montana soil I; (k) 2711, Montana soil II; and (l) 8704, Buffalo River sediment.

Sample Handling

All laboratory and field procedures and manipulations were carried out while wearing clean nitrile gloves (Kimberly-Clark, Roswell, GA). In order to prevent cross-contamination, gloves were changed frequently, and at least each time a new sample was tested.

For preparation of the colorimetric indicator solution, 0.135 g of sodium rhodizonate was dissolved in 105 mL of very cold (≈2°C) deionized water within a spray bottle, and was kept on ice to prevent deterioration of the rhodizonate solution. A fresh indicator solution was prepared daily. Vinegar extraction solution was contained in a separate spray bottle, but was not refrigerated.

In order to prevent biasing the results, tests were carried out blind by the investigator performing the colorimetric wipe procedure for lead. That is, samples were prepared for testing by a different person from the one who actually carried out the tests on the samples.

Single-element standard solutions of Ag, Al, Ba, Bi, Ca, Cd, Fe, Na, Pb, Sn, Te, and Tl (Inorganic Ventures; metal concentrations ≈1000 μg/mL) were diluted 10-fold in order to prepare solutions containing lead and/or potential interferants at desired concentrations. Also, two multielement standard solutions (Inorganic Ventures) were similarly diluted. The compositions of the multielement standard solutions were as follows: (Solution A) Sb at 500 μg/mL, Te and W at 250 μg/mL, Sn and Ti at 125 μg/mL, and Mo and Zr at 50 μg/mL; (Solution B) K at 3750 μg/mL, Cu at 2500 μg/mL, Zn at 750 μg/mL, As at 500 μg/mL, Se at 375 μg/mL, Al, Cu, Fe, Pb, Mg, P, and Ti at 250 μg/mL, La, Ag,

and V at 100 μg/mL, and Ba, Be, Cd, Cr, Co, Li, Mn, Ni, Sr, and Y at 50 μg/mL. Aliquots of the diluted solutions (500 μL) were spiked onto clean watch glasses or Petri dishes and were allowed to dry overnight.

NIST SRMs were weighed to $\pm$0.0001 g on an analytical balance (Mettler model AE163, Greifensee, Switzerland). For each SRM, a weighed amount of sample ($\approx$50 mg) was deposited lightly and uniformly on a sheet of wax paper within a 10 $\times$ 10 cm area (demarcated by a plastic template). A newly opened wipe was then unfolded and used to quantitatively collect the SRM onto the wipe surface [16]. The wipe was then laid face-up on the wax paper surface, with the collected material exposed. In order to leach lead in the collected sample, a few sprays (3–5) of vinegar were applied to the exposed wipe. After a few seconds, the wipe was then sprayed with 2–4 sprays of the rhodizonic acid indicator solution. Observation of the characteristic color change (from yellow/orange to pink/red) on the wipe surface was recorded as a positive result, while absence of this observation was recorded as a negative. Watch glasses and Petri dishes that were spiked with standard solutions and then dried were similarly tested.

Field Samples

Dermal wipe samples were collected and tested qualitatively for the presence of lead at field sites using commercial colorimetric kits ("Full Disclosure," SKC, Inc., Eighty Four, PA) in accordance with NIOSH method 9105 [8]. Dermal wipe sampling and qualitative lead testing was carried out at a metal cutting/brazing operation in northwestern Ohio (United States) and at an outdoor firing range in southwestern Ohio (United States). After colorimetric testing, the wipe samples were placed into 50 mL plastic centrifuge tubes (Becton-Dickinson, Franklin Lakes, NJ), and securely capped. The samples were then transported to the laboratory following standard chain-of-custody procedures [17]. In the laboratory, lead in these wipe samples was extracted by means of strong acid hot plate digestion (Lindberg, Thermo-Fisher Scientific, Pittsburgh, PA) following the procedure described in ASTM 1644 [18]. Subsequently, lead in wipe sample extracts was determined using inductively coupled plasma-atomic emission spectrometry (Spectro EOP, SpectroAnalytical, Fitchburg, MA) in accordance with ASTM E1613 [19].

Results and Discussion

Results from use of the lead handwipe disclosure method on $\approx$50 mg of various lead compounds are shown in Table 1. The observed intensity of the characteristic red color bloom varied for different lead compounds, and this was generally related to the solubility of each compound in aqueous solution [20]. Because lead chromate is insoluble in weak acid solution, a false negative result was observed for this matrix. For compounds giving intense positive responses, it was possible to detect the characteristic pink or red color due to the lead-rhodizonate complex for very small amounts of material (as low as $\approx$1 mg).

TABLE 1—*Qualitative colorimetric screening results from using the handwipe method on ≈50 mg of various lead compounds.*

Compound(s)	Test Response[a]
$PbCrO_4$	−
PbS, PbO_2	+
Pb metal, Pb_3O_4	++
$Pb(CH_3COO)_2$, $Pb(NO_3)_2$, PbO, $PbCl_2$, $PbBr_2$, $PbSO_4$	+++

[a](−: negative; +: weak positive; ++: definite positive; +++ brilliant positive).

With such materials, the effective identification limit for lead is apparently established by what can be discerned by the human eye [21].

Qualitative results from a number of non-lead compounds are shown in Table 2. As can be seen, it was observed that several other metals (besides Pb) gave positive responses with sodium rhodizonate. Many of these potential positive interferences, e.g., from Cd^{2+} and Hg^{+}, have been identified previously; in fact, sodium rhodizonate has been used for decades as a qualitative test for Ag^{+}, Ba^{2+}, Ca^{2+}, Sr^{2+}, and Tl^{+} [22]. However, the sensitivity of the reaction with some of these other metals is less than with Pb^{2+} [10]. Nevertheless, observation of false positives underscores one of the limitations of using qualitative screening methods.

It was desired to investigate the use of the handwipe colorimetric lead screening test on lesser masses of metals than can be investigated by weighing bulk pure materials. Therefore, it was necessary to prepare test surfaces of desired metal masses from standard solutions having target metal concentrations (as described in the experimental section). Spiked masses on watch glasses or Petri dishes ranged from ≈5 μg to ≈500 μg, depending on the metal(s) and applicable concentration ratio(s). Results from these trials are illustrated in Table 3. It is seen that Ba, Cd, and Ag give false positive results for Pb, while Fe causes a negative interference, as evidenced by a false negative. Other Pb/metal mixtures are unaffected by potentially interfering coexisting metals.

TABLE 2—*Qualitative colorimetric screening results from using the handwipe method on ≈50 mg of various non-lead compounds.*

Compounds	Test Response[a]
$Al(NO_3)_3$, Al_2O_3, Fe_2O_3, HgCl, KCl, K_2CrO_4, K_2SO_4, KNO_3, $Mg(NO_3)_2$	−
AgCl, $Bi(NO_3)_3$, $Cd(CH_3COO)_2$, $Hg(NO_3)_2$, NaCl	+
$AgNO_3$, $BaCO_3$, $BaCl_2$, $Ca(CH_3COO)_2$, $Ca(NO_3)_2$, $CdCl_2$, $Cd(NO_3)_2$, $HgCl_2$, $Zn(CH_3COO)_2$	++
$Sr(NO_3)_2$, $Ba(NO_3)_2$	+++

[a](−: negative; +: weak positive; ++: definite positive; +++ brilliant positive).

TABLE 3—*Qualitative colorimetric screening results from using the handwipe method on smooth surfaces (watch glasses or Petri dishes) prepared from dried test solutions.*

Test Metal or Mixture[a]	Test Response[b]
Mixture A; Al; Ca; Fe; K; Na; Sn; Te; Zn; Pb/Fe	−
Ba; Cd; Pb/Al; Pb/Ba; Pb/Ca; Pb/Na; Pb/Sn; Pb/Te	+
Mixture B; Ag; Pb; Pb/Ag; Pb/Cd	++

[a](1) Test surfaces of mixtures A (no Pb) and B (≈25 μg Pb) were prepared from the corresponding multielement solutions A and B described in the experimental section. (2) Metal masses ≈50 μg for single element spikes. (3) For binary (Pb/M) mixtures, M mass ≈500 μg and Pb mass ≈50 μg.
[b](−: negative; +: weak positive; ++: definite positive).

To examine data from CRMs, results from using the handwipe colorimetric lead screening test on various NIST SRMs are given in Table 4. With the exception of SRM 2709, results are negative for SRMs having lead concentrations below ≈400 μg/g. The (albeit weak) false positive result from SRM 2709 is likely due to a colorimetric reaction of rhodizonate with barium, which is present in this material at a level of nearly 1000 μg/g. At and above ≈450 μg Pb/g (≈20 μg Pb/wipe), all test results (save one, i.e., that for SRM 2711) are positive, although the intensity of the positive response varies appreciably with the matrix. For materials giving positive responses, the intensity of the characteristic

TABLE 4—*Qualitative colorimetric screening results from using the lead handwipe disclosure method on ≈50 mg of various representative NIST SRMs.*

SRM Number and Matrix[a]	SRM Pb Content (μg/g)	Wipe Pb Content (μg)	Test Response[b]
1579a (paint)	11 900	595	+++
2584 (dust)	9761	488	++
2710 (soil)	5532	277	+
2580 (paint)	4340	217	+++
2711 (soil)	1162	58.1	−
1648 (particulate matter)	655	32.8	+
2581 (paint)	449	22.5	+
2582 (paint)	208.8	10.4	−
8704 (sediment)	150	7.5	−
2583 (dust)	85.9	4.3	−
1633b (fly ash)	68.2	3.4	−
2709 (soil)	18.9	0.95	+

[a]1579a: Lead-based paint; 1633b: Coal fly ash; 1648: Urban particulate matter; 2580: Powdered paint I; 2581: Powdered paint II; 2582: Powdered paint III; 2583: Trace elements in indoor dust I; 2584: Trace elements in indoor dust II; 2709: San Joaquin soil; 2710: Montana soil I; 2711: Montana soil II; 8704: Buffalo River sediment.
[b](−: negative; +: weak positive; ++: definite positive; +++ brilliant positive).

TABLE 5—*Summary of qualitative colorimetric screening results (n = 130) obtained using the handwipe disclosing method on dermal samples tested at field sites, versus Pb mass ranges obtained from quantitative laboratory analysis.*

	Number of Test Responses[a]				
Range (μg Pb/Sample)	(−)	(+)	(++)	Percent (−)	Percent (++)
<5	39	6	1	85	2
5–10	19	6	1	73	4
10–20	13	5	5	57	22
20–50	6	5	3	43	21
>50	1	1	19	5	90

[a](−: negative; +: weak positive; ++: definite positive; note that only negatives and definite positives are used to compute percents of "meaningful" responses).

color bloom was generally greater for paint samples versus soils and sediments. This is probably due to solubility differences of the lead compounds extant in the various media.

Results from field studies of the colorimetric screening test, as applied to dermal wipe samples ($n = 130$, with similar numbers of samples from each site), are summarized in Table 5. Comparable results were observed from each of the two field sites, both in terms of ranges of lead concentrations obtained and colorimetric responses; thus, the data from the two sites were grouped together. It can be seen from Table 5 that negatives or weak positives are prevalent below about 10 μg Pb/sample, while definitive positive results predominate above 50 μg Pb/wipe. A significant overlap of positive and negative results is obtained for masses between 10 and 50 μg Pb/sample, which is consistent with what is to be expected from performance curves for qualitative test methods [13]. These data suggest that the handwipe disclosing method for lead may be useful for screening of dermal samples for this element at levels below 10 μg (for absence of Pb) and above 50 μg (for presence of Pb) per wipe. However, more study is needed in order to obtain sufficient data to accurately model the performance curve of the test method [14], and thereby obtain reliable estimates of the method's performance parameters.

Conclusions

The results of this work have highlighted some of the attributes and limitations of this qualitative screening method. Certain test matrices are obviously problematic, and false positive/false negative rates need to be investigated for each matrix of interest. For some sample matrices, the method is very sensitive; for instance, less than 20 μg Pb was detected in wipe samples collected at a firing range. However, some of the limitations of the handwipe method are obvious; thus, previous knowledge of the test matrix is highly recommended if the procedure is used for screening purposes.

A few potential applications of the handwipe disclosing method for lead can be predicted from this preliminary work. The method was effective for the detection of lead in many lead compounds employed in industry, such as oxides of lead and lead salts. Hence, the use of the handwipe method to detect appreciable levels of lead on workers' hands and on surfaces in affected workplaces is especially appealing. The procedure also appears to be effective for detecting lead at significant concentrations in indoor dust, and might be used for risk assessment and clearance purposes. However, the performance of the method on some matrices, e.g., soils, is less effective. Nevertheless, the colorimetric handwipe disclosing procedure was designed mainly as a lead screening tool for occupational settings where lead is a contaminant in dust produced during work activities. Examples of workplace environments where the use of this screening tool may be applicable include battery manufacturing/recycling, lead-based paint abatement/renovation, and firing ranges. Indeed, the vast majority of lead compounds used in industry are soluble or sparingly soluble [23], and the colorimetric handwipe procedure is effective for detecting lead in these materials.

As this was a preliminary evaluation, it remains of interest to carry out a full validation of the handwipe screening method for lead. This should be carried out in a manner that is consistent with new standard guidelines for quality assurance of qualitative analysis [24,25]. To this end, it is recommended to carry out round-robin studies so that interlaboratory data can be compared and contrasted on representative performance evaluation materials. Also, extensive field studies are necessary in order to fully evaluate the method performance in specific matrices and workplace environments.

Disclaimers

Mention of company names or products does not constitute endorsement by the Centers for Disease Control and Prevention. The findings and conclusions in this paper are those of the authors and do not necessarily represent the views of the National Institute for Occupational Safety and Health.

Acknowledgments

The writers thank Mark Boeniger for useful discussions, and the referees for valuable suggestions. Pam Drake, Bob Streicher, and Aaron Sussell critically reviewed the draft manuscript.

References

[1] Sato, M. and Yano, E., "The Association Between Lead Contamination on the Hand and Blood Lead Concentration: A Workplace Application of the Sodium Sulphide (Na_2S) Test," *Sci. Total Environ.*, Vol. 363, 2006, pp. 107–113.

[2] Durgam, S., Aristeguieta, C., and Achutan, C., "Evaluation of Health Concerns at a Printed Circuit Board Manufacturing Plant," *National Institute for Occupational*

Safety and Health (NIOSH), Health Hazard Evaluation Report No. 2007-0201—3086, CDC/NIOSH, Cincinnati, OH, 2009.

[3] Ashley, K., "Field-Portable Methods for Monitoring Occupational Exposures to Metals," *J. Chem. Health Saf.*, Vol. 17, No. 3, 2010, pp. 22–28.

[4] Pulido, A., Ruisánchez, I., Boqué, R., and Rius, F. X., "Estimating the Uncertainty of Binary Test Results to Assess Their Compliance with Regulatory Limits," *Anal. Chim. Acta*, Vol. 455, 2002, pp. 267–275.

[5] Pulido, A., Ruisánchez, I., Boqué, R., and Rius, F. X., "Uncertainty of Results in Routine Qualitative Analysis," *Trends Analyt. Chem.*, Vol. 22, 2003, pp. 647–654.

[6] Ellison, S. L. R., Gregory, S., and Hardcastle, W. A., "Quantifying Uncertainty in Qualitative Analysis," Analyst *(Cambridge, U.K.)*, Vol. 123, 1998, pp. 1155–1161.

[7] Esswein, E. J., Boeniger, M. F., and Ashley, K., "Handwipe Disclosing Method for the Presence of Lead," U.S. Patent No. 6,248,593 (2001).

[8] Esswein, E. J. and Ashley, K., "Lead in Dust Wipes by Chemical Spot Test, NIOSH Method 9105," *NIOSH Manual of Analytical Methods*, 4th ed., 3rd. suppl., CDC/NIOSH, Cincinnati, OH, 2003.

[9] Kolthoff, I. M., "Het gebruik van rhodizonzuur natrium als reagens op barium, strontium en lood [The Use of Sodium Rhodizonate as a Reagent for Barium, Strontium and Lead]," *Pharm. Weekbl*, Vol. 62, 1925, pp. 1017–1020.

[10] Feigl, F. and Suter, H. A., "Analytical Use of Sodium Rhodizonate," *Ind. Eng. Chem. Anal Ed.*, Vol. 14, 1942, pp. 840–842.

[11] Ashley, K., Fischbach, T. J., and Song, R., "Evaluation of a Chemical Spot-Test Kit for the Detection of Airborne Particulate Lead in the Workplace," *Am. Ind. Hyg. Assoc. J.*, Vol. 57, 1996, pp. 161–165.

[12] Ashley, K., Hunter, M., Tait, L. H., Dozier, J., Seaman, J. L., and Berry, P. F., "Field Investigation of On-Site Techniques for the Measurement of Lead in Paint Films," *Field Anal. Chem. Technol.*, Vol. 2, 1998, pp. 39–50.

[13] Song, R., Schlecht, P. C., and Ashley, K., "Field Screening Test Methods: Performance Criteria and Performance Characteristics," *J. Hazard. Mater.*, Vol. 83, 2001, pp. 29–39.

[14] Ashley, K., Song, S., and Schlecht, P. C., "Performance Criteria and Characteristics of Field Screening Test Methods," *Am. Lab. (Shelton, Conn.)*, Vol. 34, No. 12, 2002, pp. 32–39.

[15] ASTM E1792, 2003, "Standard Specification for Wipe Sampling Materials for Lead in Surface Dust," *Annual Book of ASTM Standards*, Vol. 04.11, ASTM International, West Conshohocken, PA.

[16] ASTM E1728, 2002, "Standard Practice for Field Collection of Settled Dust Samples using Wipe Sampling Methods for Subsequent Lead Determination," *Annual Book of ASTM Standards*, Vol. 04.11, ASTM International, West Conshohocken, PA.

[17] ASTM D4840, 1999, "Standard Guide for Sampling Chain-of-Custody Procedures," *Annual Book of ASTM Standards*, Vol. 11.01, ASTM International, West Conshohocken, PA.

[18] ASTM E1644, 2004, "Standard Practice for Hot Plate Digestion of Dust Wipe Samples for the Determination of Lead," *Annual Book of ASTM Standards*, Vol. 04.11, ASTM International, West Conshohocken, PA.

[19] ASTM E1613, 2004, "Standard Test Method for Determination of Lead by Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-AES), Flame Atomic Absorption Spectrometry (FAAS), or Graphite Furnace Atomic Absorption Spectrometry (GFAAS) Techniques," *Annual Book of ASTM Standards*, Vol. 04.11, ASTM International, West Conshohocken, PA.

[20] Merck, *The Merck Index*, 11th. ed., S. Budavari, Ed., Merck, Rahway, NJ, 1989.

[21] Note: Red-green color-blind individuals may have difficulty discerning this color change.
[22] Feigl, F. and Anger, V., *Spot Tests in Inorganic Analysis*, 6th ed., Elsevier, Amsterdam, 1972.
[23] Greninger, D., Kollonitsch, V., Kline, C. H., Willemsens, L. C., and Cole, J. F., *Lead Chemicals*, International Lead Zinc Research Organization, New York, 1975.
[24] Ríos, A., Barceló, D., Buydens, L., Cárdenas, S., Heydorn, K., Karlberg, B., Klemm, K., Lendl, B., Milman, B., Niedhart, B., Stephany, E. W., Townshend, A., Zschunke, A., and Valcárcel, M., "Quality Assurance of Qualitative Analysis in the Framework of the European Project 'MEQUALAN'," *Accreditation Qual. Assur*, Vol. 8, 2003, pp. 68–77.
[25] Trullols, E., Ruisánchez, I., and Rius, F. X., "Validation of Qualitative Analytical Methods," *Trends Analyt. Chem.*, Vol. 23, 2004, pp. 137–145.

Reprinted from JAI, Vol. 8, No. 5
doi:10.1520/JAI103527
Available online at www.astm.org/JAI

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Handwipe Method for Removing Lead from Skin

ABSTRACT: Researchers at the U.S. National Institute for Occupational Safety and Health (NIOSH) developed a handwipe removal method for lead (Pb) after field studies showed that workers in lead-acid battery plants had significant risks for dermal-oral lead exposures, despite their attempts to remove the lead by washing with soap and water. Hand washing with soap and water remains the standard recommendation for workers (as well as the public) to clean skin known or believed to be contaminated with toxic metals, such as lead. Despite longstanding recommendations for workers to "wash hands with soap and water," no efficacy studies show this to be a completely effective removal method for lead. Removal of toxic metals such as lead from skin constitutes a decontamination procedure; it is not, in fact, a hand-washing step. NIOSH scientists conceived and developed a highly effective (nearly 100 %) method for removal of lead from skin. A systems approach was devised incorporating four components deemed necessary for effective metal removal: Surfaction, *p*H control, chelation, and mechanical effects. The handwipe removal method evolved from a previous NIOSH invention, the handwipe disclosing method for the presence of lead, in the interests of providing complementary techniques for dermal lead detection and

Manuscript received October 29, 2010; accepted for publication March 21, 2011; published online May 2011.

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Presented at the ASTM International Symposium on Surface and Dermal Sampling, October 14–15, 2010, San Antonio, TX. This article was prepared by U.S. Government employees as part of their official duties and legally may not be copyrighted in the United States of America.

Cite as: Esswein, E. J., Boeniger, M. F. and Ashley, K., "Handwipe Method for Removing Lead from Skin," *J. ASTM Intl.*, Vol. 8, No. 5. doi:10.1520/JAI103527.

decontamination. The method is a patented, award-winning, commercialized technology that has significant potential to prevent occupational and public exposures to lead.

KEYWORDS: isostearamidopropyl morpholine lactate (ISML), cleanser, citric acid, decontamination, dermal, lead, wipe, workplace

Introduction

Occupational and environmental exposure to metals (e.g., lead [1]) and other elements inherently toxic to biological systems (e.g., cadmium, arsenic, beryllium) implies (depending on the degree of exposure) a potential for adverse effects on workplace and public health. Exposure to such metals, especially lead, is a significant problem that affects a large and diverse segment of the population, and workers and their families are especially at risk [2–4]. Exposure to lead (Pb) may occur in a wide variety of locations, including the workplace, homes or schools, or the outdoor environment [1,2,4]. Skin contact is a significant route for transfer, and exposure to metals such as lead. While hand-to-mouth transfer is understood to be the most significant route of exposure; (a) several researchers have shown that lead ions may be absorbed through the skin [5–7]; (b) skin can act as a reservoir for metals [8,9]; (c) skin surface deposition can be an important source of secondary contamination [3,8,10]; and (d) impairment or loss of skin barrier function can occur [6,11]. Additionally, skin contact with some metals and their compounds (nickel, chromium, and beryllium among others) can cause sensitization and systemic allergic responses which can result in serious occupational disease and even loss of workers from the working population [12]. Unfortunately, many toxic metals are not easily washed off of the skin; finely divided metal particles not only lodge within the complex interstices of the stratum corneum but also bind to sulfhydryl, carboxyl, and other groups present in skin proteins [11]. Industrial hygiene workplace investigations conducted by NIOSH and other investigators have shown that lead and other metals remain on the hands of workers even after they report, or were known to have washed their hands before eating [13–16]. In industrial settings where lead poses exposure risks, significant metal contamination may remain even after washing [13,14,16,17].

Lead provides a particularly useful illustration of the exposure risks, detection and decontamination challenges posed by skin exposures. Lead exposure can occur to workers (and the public) during and after removal of lead-based paints and/or the renovation of structures containing lead-based paints [2], to workers in waste-to-energy plants, manufacture of lead-acid batteries, and other related industries (e.g., radiator repair work, welding, and construction work). Adults or children living within or visiting homes or schools containing deteriorated lead-based paints can be at risk for exposures [4,10]. Lead residues on the skin, especially on the hands, of industrial workers can be a significant health risk since such residues can be invisible and may be ingested during normal activities (e.g., eating, drinking, and smoking) [7–9,17,18]. Contaminated clothing (as well as automobiles) presents take-home

toxics issues for workers and their families; lead is an especially important metal in this route of exposure [19].

Although screening methods for detecting the presence of lead in workplaces are available [20–22], validated methods, techniques, and products for highly effective (>99%) skin and surface decontamination are needed. In response to this gap, efforts have been directed to acknowledge the need and importance of detecting and removing lead from skin and other surfaces [6,7,23]. Lead desorption and removal from contaminated soil using surfactants has been investigated and described [24,25]. The physical properties of liquids and soil permit physical mixing of contaminated liquids or soils with decontaminating agents (such as surfactants) on a microscopic level that cannot be achieved with non-dispersible (such as, solid) matter. As a result, it may be more difficult to contact and remove metal contamination from a solid surface, especially where the surface has interstices where contaminants can lodge and bind (such as skin). Moreover, agents suitable for lead decontamination of liquids or soils may cause damage to solid surfaces and, in particular, may irritate or harm sensitive surfaces such as human skin [6,26,27].

Products that claim to remove lead and toxic metals from human skin often contain active ingredients such as the chelating agent ethylenediaminetetraaceticacid (EDTA) or anionic surfactants (fatty acid soaps) [28]. EDTA, while a good chelating agent, is a suspected persistent environmental pollutant and a skin irritant. EDTA may cause reddening or inflammation on prolonged skin contact [29]. Anionic surfactants such as sodium laurel sulfate (SLS) are used in many surfactant or soap mixtures but SLS may also cause skin irritation, including dryness and scaling [26,30]. Moreover, anionic surfactants due to their alkaline *p*H may not be fully effective in mobilizing and removing lead contamination [8,24,25,31]. Therefore, considering the disadvantages of EDTA and anionic surfactants for removing metals from human skin and surfaces, safe, reliable and effective compositions are needed for removing metals, notably lead. Of particular need are compositions and methods that do not substantially damage the treated surface, or unduly irritate, or sensitize biological surfaces, notably skin [32].

In this work we describe the development and evaluation of treated wipes for decontamination of lead and other metals from surfaces such as skin. Optimal wipe materials were found to be those that included a three-dimensionally highly textured absorbent support such a creped surface (a textured surface comprising a succession of ridges and groves; see Fig. 1) with isostearamidopropyl morpholine lactate (ISML) and citric acid in the absorbent, creped wipe. The amounts of ISML and citrate in the wipe medium were optimized in order to obtain the best metal-removing capability from dermal surfaces. The metal-removal performance of these wipes from the hands of the researchers as well as volunteers was compared to that of several other commercially available products and formulations, and also to hand washing with soap and water. We report herein the results of these investigations, which demonstrate the superior decontamination effectiveness of a textured (creped) wipe substrate containing the cationic surfactant ISML and chelating and *p*H adjusting agent, citric acid.

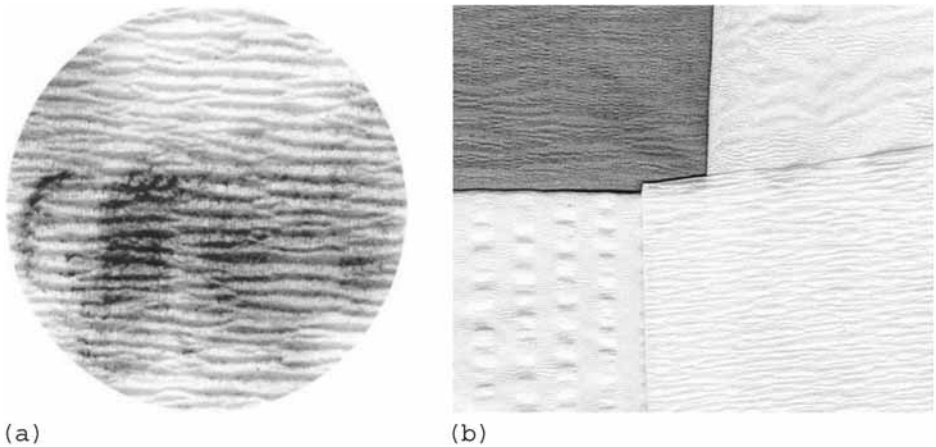

FIG. 1—*Creped wipe materials (a) Photo of creped wipe showing succession of ridges and groves and sample collection within a textured wipe. Photo courtesy of Micrex Corporation. Used with permission B) Photo of various creped textures that can be imparted into wiping materials. Photo courtesy of Micrex Corporation. Used with permission.*

Experimental

Materials

Initial formulations of the decontamination wipes were made in NIOSH laboratories in Cincinnati, OH and Denver, CO. Several rolls of DuPont™ Sontara® were creped by Micrex® (Walpole MA) and provided to NIOSH as the absorbent substrate for the initial experimental trials. ISML (Mackalene™ 426 and Incromate® ISML) were obtained from McIntyre Group Ltd. (University Park, IL USA) and CRODA Inc. (Edison, NJ, USA), respectively. Citric acid solutions were made in-house (NIOSH, Cincinnati, OH, USA). A pre-commercial version of the invention (MEDTOX® Wipe Lot # 0807) of LeadTech™ Wipes was provided for evaluation by MEDTOX® Diagnostics (Burlington, NC, USA). The final formulation of wipes contained between 0.3 and 2 g (g) of ISML and between 0.01 and 0.1 g citric acid per gram of absorbent support (exact formulations are proprietary). Sampling wipes (Palintest™ and Ghost Wipes™) meeting international voluntary consensus performance standards were obtained from Palintest® (Gateshead, Tyne and Wear, U.K.) and Environmental Express (Mt. Pleasant, SC, USA), respectively [33]. D-Lead™ cleansers, both with and without scrubbers, were obtained from Esca Tech (Milwaukee, WI, USA). Clean-All Heavy Metals Soap™ cleanser came from Sasha International (Miami Beach, FL, USA). Kresto Select™ cleanser with scrubber, and Kresto Kwik™ wipes, were purchased from Stockhausen (Greensboro, NC, USA). GoJo Multigreen™ cleanser with scrubbers was obtained from GoJo Corp. (Akron, OH, USA). Waxed kitchen paper, paper towels, and food grade corn starch (Safeway

brands, Pleasanton, CA, USA), as well as Pampers Baby Wipes™ and Ivory™ Liquid Soap (Procter & Gamble Co., Cincinnati, OH, USA), were purchased at a neighborhood grocery store. Disposable nitrile laboratory gloves came from Fisher Scientific (Fair Lawn, NJ, USA). Acetic acid (reagent grade) came from Sigma Aldrich. Industrial hand soap used was Smart & Final™ Liquid Soap (Los Angeles, CA, USA).

Red lead monoxide powder (PbO; $>$ 99.9 %, particle size $<$10 micrometer[μm]) was obtained from Sigma-Aldrich (Milwaukee, WI, USA) and was mixed uniformly into corn starch (used as a mixing diluent) by rotary tumbling to yield a concentration of 90.9 mg (mg) Pb/gram. Aliquots of this mixture were weighed into samples of 33 mg each (to $\pm$1 mg) on an analytical balance (Mettler model AE163, Greifensee, Switzerland). Lead-containing dust (consisting primarily of PbO collected from a lead-acid battery manufacturing plant in Texas, USA [14]) was also weighed into samples of approximately 3000 μg each. Polyethylene centrifuge tubes (Elkay™, 50 mL) were obtained from Life Sciences Products (Denver, CO, USA).

Procedures

The evaluation of lead dust decontamination from human hands (the researchers as well as volunteers) was approved through NIOSH human subjects review board [34]. Skin was contaminated by spiking both palmer surfaces with weighed quantities of leaded dust (either lead monoxide powder and corn starch, or straight PbO from a lead-acid battery plant). Skin sampling was performed in accordance with NIOSH Method 9105 [35]. To establish if significant lead might have been present as background contamination, an initial 30-s hand wipe sample was collected on every subject (researcher or volunteer) before each of the experimental trials. Both Ghost Wipes™ and Palintest™ brand wipes were used for skin sampling. To collect skin samples for the presence of lead, the investigator (wearing clean nitrile gloves) opened a wipe packet and offered the folded wipe to the subject. The volunteer was asked to completely unfold the wipe and then carefully wipe to sample the palmer surfaces of both hands for 30 s to collect an initial background sample for the presence of lead. After 30 s, the subject was requested to stop wiping and fold the wipe with the soiled surface facing inward. The volunteer was asked to place the folded wipe into a 50-mL plastic centrifuge tube that was used for sample containment and laboratory transport. After sample collection, the centrifuge tube was tightly capped and labeled with a discrete sample identification number using an indelible marker.

To apply the leaded dust to the skin of each volunteer, the investigator (wearing clean nitrile gloves) carefully unfolded the sample weighing paper and poured each pre-weighed leaded dust sample into the volunteers' cupped hands while they were held over a clean sheet of wax paper. The wax paper was placed below the subjects' hands to capture any leaded dust that fell off the hands during the application, enabling a mass balance to be established. Subject individuals were asked to carefully rub the leaded

dust into the skin of their hands for 30 s, being careful to keep as much lead dust on their hands as possible.

After the leaded dust was applied to the skin, the investigator changed gloves, removed a decontamination wipe from its container and handed it to each volunteer. Each subject was asked to cleanse his/her hands for a period of 30 s. Following the 30-s decontamination step, the volunteer was asked to rinse their hands for 30 s under a laboratory sink with flowing, tepid water to remove the surfactant. After rinsing, the investigator gave the volunteer two flat paper towels and instructed the volunteer to carefully pat dry their hands, taking care not to rub their hands with the paper towel.

Two serial handwipe samples were then collected to evaluate the volunteers' (or researchers') hands for the presence of lead on skin. The investigator (wearing a fresh pair of nitrile gloves) again opened packets of Ghost Wipes™ or Palintest™ wipes and offered the folded wipes to the volunteers. The volunteers were asked to unfold the wipe and wipe the palmer surfaces of both hands for 30 s and then to fold the wipe together with the "soiled" or sample side facing in. The volunteers placed the wipes into 50-mL centrifuge tubes, which were then capped and labeled. Skin sampling was repeated twice and the samples combined in a single tube for analysis. A surface wipe sample was collected from the wax paper that was placed on the laboratory bench below the volunteers' hands during application of leaded dust to the hands. This sample was used to account for any lead-containing dust that might have not been rubbed into the volunteers' hands or somehow spilled through their fingers during the application process.

Similar protocols as described above were employed to evaluate hand washing with soap and water as well as various liquid soaps, solutions, wipes and cleanser formulations.

Wipe samples were analyzed at Bureau Veritas North America, (Novi, MI, USA), a facility accredited by the American Industrial Hygiene Association Laboratory Accreditation Programs, LLC. Analyses were carried out using NIOSH method 9102 with modifications: lead in collected wipe samples was determined by means of nitric/perchloric acid hot block digestion and inductively coupled plasma-atomic emission spectrometry (ICP-AES). Each wipe sample was removed from the centrifuge tubes and placed in a clean beaker to which 2.5 mL of 12.1*M* perchloric acid was added and allowed to stand for 30 min. The beakers were placed in a hot block and heated at 95°C for 15 min. Samples were removed from the hot block, left to stand, and allowed to cool to room temperature, and 2.5 mL of 15.6*M* nitric acid was then added. The samples were placed back in the hot block and again heated at 95°C for 15 min. The samples were then removed, left to stand, and allowed to cool to room temperature and diluted to a final volume of 25 mL with deionized water. Quality assurance/quality control samples (blank samples, spikes and spike duplicates) were digested and analyzed in the same manner. All samples were analyzed using a Perkin Elmer Optima 3200 XL ICP-AES instrument (Boston, MA, USA). The ICP-AES limits of detection and quantitation for lead (0.3 and 0.86 μg per sample, respectively) were estimated in accordance with ASTM E1613 [36].

Results and Discussion

In initial experiments, the researchers compared the efficacy of a combination of liquid surfactant and acids alone, with no wipe (i.e., ISML and acetic or citric acids) against ISML and citric acid added to a lightly textured "creped" wipe, against common industrial soap and water to remove leaded dust from the researcher's skin. Fig. 2 shows the amount of lead (in μg) remaining on the palmer surfaces of the skin after an initial 3000 μg Pb loading (using the PbO–corn starch mixture) following the four different methods of cleansing: (1) Common industrial soap and water alone (S&W); (2) liquid only mixtures of citric acid and ISML (C-I liquid); (3) liquid mixtures of acetic acid and ISML (A-I liquid); and, (4) an aqueous mixture of citric acid and ISML (C-I on wipe) applied onto a commercially available lightly textured (Pampers® brand) baby wipe. In these trials, five replicates were run for each of the above four experiments.

As is illustrated in Fig. 2, use of common industrial soap and water alone was not effective in completely removing deposited lead from human skin, as nearly 300 μg of lead remained after hand washing. The alkaline (pH 8–9) nature of common industrial soap and water, absence of a low pH surfactant and chelating agent, and lack of mechanical removal effects are understood to be the main reasons for less complete removal of lead from skin. In contrast, a citric acid and ISML formulation on a lightly textured "creped" wet wipe was the most effective in this trial, as evidence by the least amount of remaining lead recovered (< 75 μg) after skin decontamination. Mixtures of citric or acetic acids and ISML applied without a wipe (C-I liquid and A-I liquid) were also

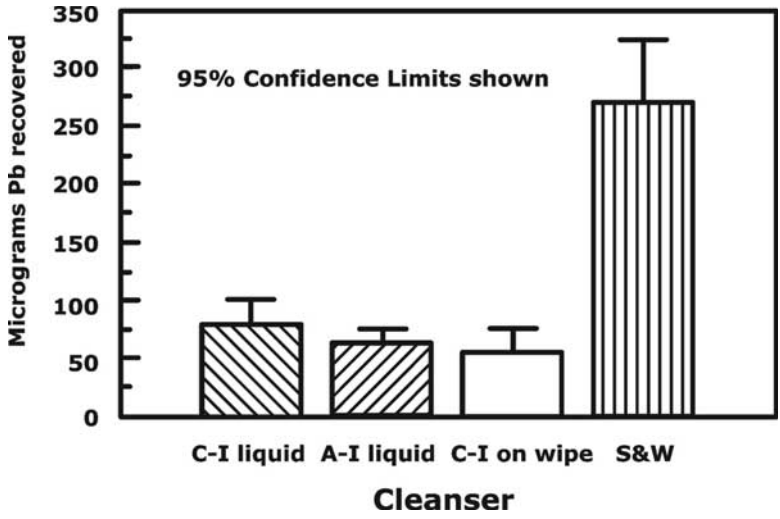

FIG. 2—*Amount of lead (in μg) remaining on human hands after cleansing with a liquid mixture of citric acid and ISML ("C-I liquid"), a liquid mixture of acetic acid and ISML ("A-I liquid"), a mixture of citric acid and ISML on a wipe ("C-I on wipe"), and plain soap and water ("S&W"); n=5 for each cleanser method.*

effective at removing lead (Fig. 1). The finding that the wipe formulation with citric acid and ISML is somewhat more effective than ISML/citric acid liquid only indicates that the mechanical action of even a lightly textured wiping material contributed to the lead-removal process.

In another initial set of experiments also conducted on the researchers' hands, several variations on the use of a highly textured "creped" wipe containing aqueous solutions of citric acid and ISML were tested for efficacy at removing lead from skin. Different cleansing protocols utilizing Micrex® (highly textured, creped) Dupont Sontara wiping material were employed after application of 3000 μg Pb in leaded dust on hands, as described previously. These six protocols and hand washing with common soap and water (which is the most commonly used cleansing protocol) are outlined in Table 1. Each protocol was done in replicates of five.

As shown in Fig. 3, Protocols A and B (using two different concentrations of citric acid and ISML) were essentially equally effective at removing lead from skin, with $\approx$15 μg Pb of an initial 3000 μg Pb load measured on the hands after cleansing in this manner. With Protocol C, (same as A&B but without a water rinse) about 100 μg Pb remained on the hands, indicating that a final water rinse is recommended for removal of solubilized lead. Protocol D (use of a second citrate/ISML creped wipe) illustrates that use of a second citric acid and ISML treated wipe removes almost all lead, with only 2.5 μg Pb remaining on skin. Use of a second wipe but without a final water rinse (Protocol T) indicates that lead removal is effective ($\approx$35 μg Pb remains after the cleansing protocol). Protocol T could be used in remote locations where water for rinsing is not available.

In contrast to each of the protocols involving one or more citrate/ISML wipes, the use of a wetted, creped wipe with no surfactant or citric acid (i.e., Protocol E) was less effective for lead removal from skin, with nearly 200 μg Pb remaining (see Fig. 3) indicating that the mechanical action of the creped wipe has an effect in dislodging lead from skin. Use of common industrial hand soap

TABLE 1—*Cleansing protocols tested in evaluating efficacies of treated, textured wipes.*

Protocol	Description
A	Wipe containing 10 mL of 0.5 % citric acid and 12.5 % ISML solution and a final water rinse
B	Wipe containing 10 mL of 0.25 % citric acid and 18.75 % ISML solution with a final water rinse
C	Protocol B but without a final water rinse
D	Protocol B but with use of a second wipe prior to rinsing with water
E	Use of a wipe wetted with water only, followed by a water rinse
T	Protocol B followed by a second citric acid/ISML wipe, but with no water rinse
S&W	Soap and water hand washing

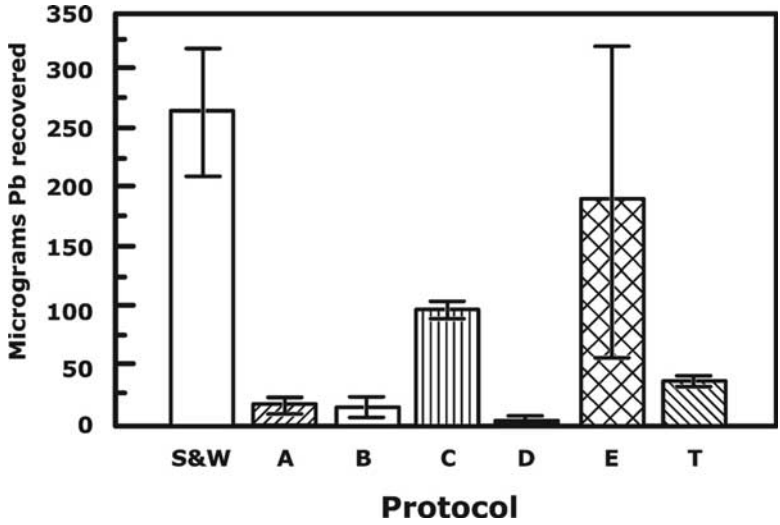

FIG. 3—*Amount of lead (in μg) remaining on human hands after cleansing protocols involving one or more citrate/ISML wipes, as described in Protocols A, B, C, D and T; or a wipe wetted only with water (Protocol E); or hand washing with soap and water (S&W). 95 % confidence limits are shown (n=5). (See text and Table 1 for description).*

and water as cleaning agents (S&W) resulted in over 250 μg Pb remaining on the hands thereby demonstrating the relative ineffectiveness of this widely used method for lead removal from dermal surfaces.

The efficacy of lead removal using textured creped wipes containing citric acid and ISML was compared side-by-side with other commercial products in described in Fig. 4. In this set of experiments, Dupont Sontara™ creped by Micrex® (20 cm × 20 cm) were fortified with 10 mL of aqueous solution containing 0.5 % citric acid and 12.5 % ISML. Fourteen volunteer's hands were spiked with 3000 μg leaded dust prior to cleansing. Commercial products tested included those listed in Table 2. These comparison products were selected as among the most widely used on the commercial market for lead and other heavy metal decontamination. The scrubbers in some of these products may consist of ground walnut shells, plastic beads, or crystalline silica (comparable to the consistency of beach sand).

The citric acid/ISML wipe (Cleanser A) provided statistically significant superior lead cleansing from human hands when compared to Cleansers F, U, N, H, and G (cleansers listed in Table 2). The citric acid/ISML wipe also removed more lead from skin than did Cleanser I (16 μg Pb versus 27 μg Pb remaining on hands: Fig. 3), but this difference was not statistically significant. It is noted that scrubbers contained in some of the commercial products may irritate the skin with repeated use [32].

As a follow-on to the previous investigations, a blind comparison between citric acid/ISML wipes and representative liquid cleansers was also conducted using another fourteen volunteer participants. In this investigation, the

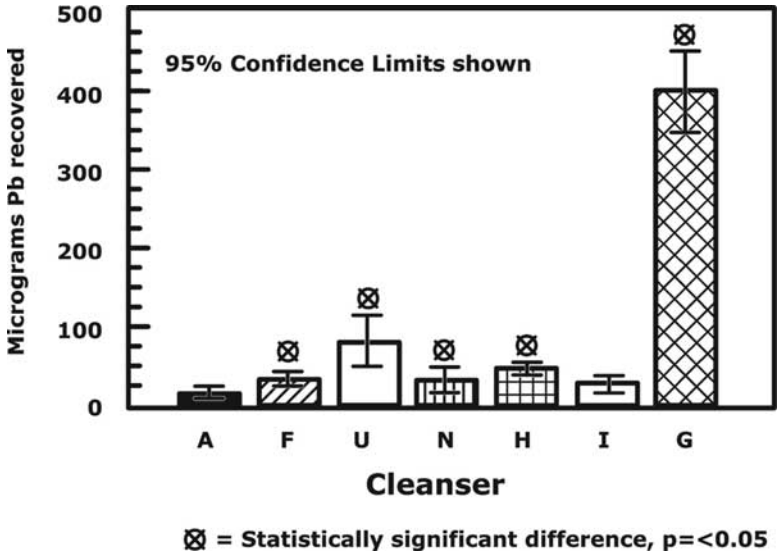

FIG. 4—*Amount of lead remaining on hands following 3000 μg Pb initial loading and after cleaning with a citric acid/ISML wipe (Cleanser A) and various commercially available industrial hand cleansers (Cleansers F, U, N, H, I and G), as described in text and Table 2; n = 5 for each cleanser method.*

participants were not informed of the identities of the test products. Each subject was provided with a randomly selected cleanser, and each product was used twice by each participant. As in previous trials, volunteers' hands were spiked with 3000 μg Pb leaded dust prior to cleansing. The liquid cleansers that were compared to the citric acid/ISML wipe (Cleanser A) included: 1. D-Lead® liquid soap without scrubbers (Cleanser B); 2. Clean-All Heavy Metal™ liquid soap (Cleanser C); and 3. Ivory® liquid soap (Cleanser D). In carrying out these tests, 2 mL of each of the liquid soaps were applied to the palms.

TABLE 2—*Cleansing products tested in comparison study with treated, textured wipes containing citric acid and ISML.*

Cleanser	Product Description
A	Wipe containing 10 mL of 0.5 % citric acid and 12.5 % ISML solution
F	Clean-All Heavy Metal Soap
G	Kresto Kwik Wipes
H	Kresto Select with scrubber
I	GoJo Multigreen with scrubbers
N	D-Lead with scrubbers
U	D-Lead without scrubbers

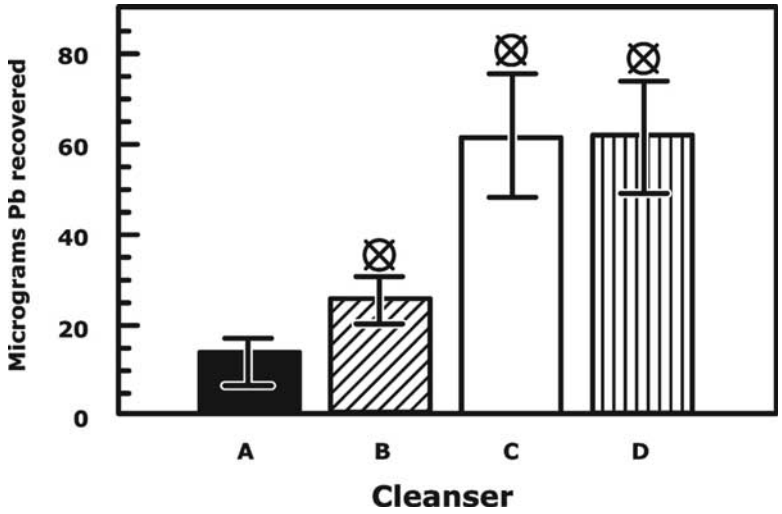

FIG. 5—*Amount of a 3000 μg Pb initial load remaining on the hands of 14 blinded study participants after using a citric acid/ISML wipe ("Cleanser A"), D-Lead liquid soap without scrubbers ("Cleanser B"), Clean-All liquid soap ("Cleanser C"), or Ivory liquid soap ("Cleanser D").*

The results from these comparisons are shown in Fig. 5. It is demonstrated that the citric acid/ISML wipe system removed more lead from the hands than each of the other products; these results are statistically significant. One of the products advertised to be effective for removal of lead as well as other toxic metals (Clean-All Heavy Metal Soap™) in fact did not decontaminate hands any better than (the non-industrial) Ivory® liquid soap.

In a final evaluation, the efficacy of lead removal using straight, lead-acid battery plant dust (as 99 % PbO), of a licensed, converted (commercially manufactured and packaged), Beta version of the invention was evaluated using nine volunteers. Results from these experiments are summarized in Table 3. The spiking, sampling, and decontamination investigative protocol was similar to the previous investigations but Palintest™, rather than Ghost Wipes™ were used for skin sampling. Differences in sampling efficiency using Ghost Wipes™ and Palintest™ brand wipes has been investigated and no significant sample collection efficiency differences were found [37]. Not unexpectedly, all background handwipe samples revealed some lead, likely from handling lead-containing brass keys, or touching other brass/lead containing environmental surfaces. Dermal lead concentrations ranged from trace levels (detectable but not quantifiable) to 2.6 μg Pb/handwipe. An average of 670 μg of lead was recovered in surface wipe samples from the wax paper suggesting the application technique varied considerably in the successful loading of lead dust onto the skin. The average calculated amount of Pb applied to the skin was $\approx 2,300$ μg. Calculated percent removal for pre-commercial lot 0807 ranged from 99.7 to 99.9 %, indicating that the MEDTOX™ Wipe (which uses a slightly

TABLE 3—*Lead dust removal efficacy (hands) from a pre-commercial lot of textured citric acid/IMSL wipes.*

Volunteer No.	Background Handwipe Pb (μg)	Initially Weighed Pb Amount (μg; as PbO)	Pb (μg) Recovered from Wax Paper	Calculated Pb Mass (μg) Applied to Hands	Final Pb (μg) Collected After 2 Serial Handwipes	Calculated % Pb Removed[a]
1	2.5	2868	830	2038	4.6	99.7
2	0.31[b]	2978	310	2668	6.6	99.7
3	2.0	3238	1500	1738	2.1	99.8
4	2.5	2830	660	2170	2.7	99.8
5	0.65[b]	3146	270	2876	3.3	99.8
6	2.6	2960	600	2360	1.8	99.9
7	0.77[b]	3006	790	2216	1.9	99.9
8	1.2	2997	470	2527	1.6	99.9
9	0.71[b]	3053	600	2453	3.5	99.8

[a]Accounts for initial hand contamination measured (background) and losses onto wax paper that occurred during loading of lead oxide dust onto hands.
[b]Estimated amount: Above detection limit but below quantitation limit.

different but highly textured wipe material from that supplied to NIOSH by Micrex®) was as effective as the original citric acid/ISML creped wipes evaluated previously (creped DuPont™ Sontara® wipe material supplied by Micrex®). Similar experiments using Hygenall® brand licensed and commercialized wipes have also demonstrated 99.2 % lead removal efficiency with five trials using 6,000 μg palmer skin loadings and of straight PbO (99 %) from a lead-acid battery plant.

Conclusion

A novel and highly effective method for removing toxic metals (notably lead) from skin has been conceived, developed, evaluated, patented and licensed from the government to the private sector. The technology consists of a three-dimensionally textured absorbent wipe treated with proportions of a cationic surfactant (ISML) and a weak organic acid (citric acid). Published research has shown that the method does not damage the skin [6]. The technology design criteria involved developing a system of metal removal incorporating contributing effects of surfaction, chelation, *p*H adjustment and mechanical removal. This technology was developed to complement a previous NIOSH invention involving colorimetric chemistry that detects lead collected from skin and workplace surfaces. Used serially, the two technologies are envisioned to "close the loop" on detection and decontamination of skin contaminated with lead. Decontamination of workers' skin should improve with the use of this technology and the commercial versions of these wipes, which have been shown to be more effective than hand washing using soap and water.

Acknowledgments

The authors would like to thank Ms. Tami Wise, NIOSH, DART for her expert assistance in preparation of a proportion of the treated wipes used in this study, and careful weighing of lead dust samples. The authors also thank Dr. Cynthia A.F. Striley, NIOSH, DART, for her assistance with resolution of graphics used in the manuscript. The authors also thank the volunteers who agreed to participate in this study. Finally, the authors thank Mr. Richard Walton of Micrex Corporation for donating creped wipe material and for consistently generous and wise council regarding all things webbed, creped and converted.

References

[1] US Agency for Toxic Substances and Disease Registry (ATSDR), *Toxicological Profile for Lead*, ATSDR, Atlanta, GA, 2007.

[2] Sussell, A., Ed., "Protecting Workers Exposed to Lead-Based Paint Hazards: A Report to Congress," *DHHS (NIOSH) Publ. No. 98-112*, CDC/NIOSH, Cincinnati, OH, 1997.

[3] Weber, A., Ed., "Health Hazard Evaluations: Issues Related to Occupational Exposure to Lead, 1994–1999," *DHHS (NIOSH) Publ. No. 2001–113*, CDC/NIOSH, Cincinnati, OH, 2001.

[4] Whelan, E. A., Piacitelli, G. M., Gerwel, B., Schnorr, T. M., Mueller, C. A., Gittleman, J., and Matte, T. D., "Elevated Blood Lead Levels in Children of Construction Workers," *Am. J. Public Health*, Vol. 87, 1997, pp. 1352–1355.

[5] Stauber, J. L., Florence, T. M., Gulson, B., and Dale, L., "Percutaneous Absorption of Inorganic Lead Compounds," *Sci. Total Environ.*, Vol. 145, 1994, pp. 55–70.

[6] Filon, F. L., Boeniger, M., Maina, G., Adami, G., Spinelli, P., and Damian, A., "Skin Absorption of Inorganic Lead (PbO) and the Effect of Skin Cleansers," *J. Occup. Environ. Med.*, Vol. 48, 2006, pp. 692–699.

[7] Sun, C.-C., Wong, T.-T., Hwang, Y.-H., Chao, K.-Y., Jee, S.-H., and Wang, J.-D., 2002, "Percuteanous Absorbtion of Inorganic Lead Compounds," *Am. Ind. Hyg. Assoc. J.*, Vol. 63, pp. 641–646.

[8] Esswein, E. and Boeniger, M., "Preventing the Toxic Hand-Off," *Occup. Hazards*, Vol. 67, 2005, pp. 53–61.

[9] Far, H. S., Pin, N. T., and Kong, C. Y., "An Evaluation of the Significance of Mouth and Hand Contamination for Lead Absorption in Lead Acid Battery Plant Workers," *Int. Arch. Occup. Environ. Health*, Vol. 64, 1993, pp. 439–443.

[10] Piacitelli, G. M., Whelan, E. A., Sieber, W. K., and Gerwel, B., "Elevated Lead Contamination in Homes of Construction Workers," *Am. Ind. Hyg. Assoc. J.*, Vol. 58, 1997, pp. 447–454.

[11] Hostynek, J., "Factors Determining Percutaneous Metal Absorption," *Food Chem. Toxicol.*, Vol. 41, 2003, pp. 327–345.

[12] Hostýnek, J., Hinz, R. S., Lorence, C. R., Price, M., and Guy, R. H., "Metals and the Skin," *Crit. Rev. Toxicol.*, Vol. 23(2), 1993, pp. 171–235.

[13] Esswein, E. J. and Tepper, A., NIOSH Health Hazard Evaluation Report No. 91-0366-2453, Delaware County Resource Recovery Facility, Chester, PA. CDC/NIOSH, Cincinnati, Ohio, 1991.

[14] Esswein, E. J. and Boeniger, M. F., NIOSH Health Hazard Evaluation Report No. 94-0268-2618, Standard Industries, San Antonio, TX, CDC/NIOSH, Cincinnati, OH, 1996.

[15] Durgam, S., Aristeguieta, C., and Achutan, C., NIOSH Health Hazard Evaluation Report No. 2007-0201-3086, Sanmina-SCI® Corporation, CDC/NIOSH, Huntsville, AL, 2009.

[16] Mattorano, D., NIOSH Health Hazard Evaluation Report No. 94-0273-2556, Bruce Mansfield Power Station, Shippingport, PA, CDC/NIOSH, Cincinnati, OH, 1996.

[17] Chavalitnitikul, C., Levin, L., and Chen, Lung-Chi, "Study and Models of Total Lead Exposures of Battery Workers," *Am. Ind. Hyg. Assoc. J.*, Vol. 45, 1984, pp. 802–808.

[18] Chuang, H.-Y., Lee, M.-L T., Chao, K.-Y., Wang, J.-D., and Hu, H., "Relationship of Blood Lead Levels to Personal Hygiene Habits in Lead Battery Workers: Taiwan, 1991–1997," *Am. J. Ind. Med.*, Vol. 35, 1999, pp. 595–603.

[19] Hipkins, K. L., Materna, B. L., Payne, S. F., and Kirsch, L. C., "Family Lead Poisoning Associated with Occupational Exposure," *Clin. Pediatr. (Phila.)*, Vol. 43, 2004, pp. 845–849.

[20] Esswein, E. J., Boeniger, M. F., and Ashley, K., "Handwipe Disclosing Method for Lead," U.S. Patent No. 6,248,593 (2001).

[21] Ashley, K., "Field-Portable Methods for Monitoring Occupational Exposures to Metals," *J. Chem. Health Saf.*, Vol. 17, 2010, pp. 22–28.

[22] 2003 U.S. National Institute for Occupational Safety and Health (NIOSH), Lead in Dust Wipes by Chemical Spot Test Method, (Colorimetric Screening Method) 9105, NIOSH Manual of Analytical Methods, CDC/NIOSH, Cincinnati, OH, www.cdc.gov/niosh/nmam (Last accessed 20 Sept. 2010).

[23] Askin, D. P. and Volkmann, M., "Effect of Personal Hygiene on Blood Lead Levels of Workers at a Lead Processing Facility," *Am. Ind. Hyg. Assoc. J.*, Vol. 58, 1997, pp. 752–753.

[24] Kornecki, T. S., Brown, G. O., Allred, B., and Basta, N., "Cationic Surfactant Feasibility for Use in Removal of Lead from Soil," *Envir. Geosciences*, Vol. 5, 1998, pp. 29–38.

[25] Kornecki, T. S., Brown, G. O., and Allred, B., "Saturated Column Feasibility Study Using Cationic Surfactants for In Situ Removal of Lead from Soil," *Envir. Geosciences*, Vol. 6(1), 1999, pp. 42–52.

[26] Froebe, C., Simion, F., Rhein, L., Cagan, R., and Kligman, A., "Stratum Corneum Lipid Removal by Surfactants: Relation to In Vivo Irritation," *Dermatologica*, Vol. 181, 1990, pp. 277–283.

[27] Schleupein, R. and Ross, L., "Effects of Surfactants and Solvents on the Permeability of the Epidermis," *J. Soc. Cosmet. Chem.* Vol. 21, 1970, pp. 853–857.

[28] 2010 Esca Tech Inc., D-Wipe®, Esca Tech, Milwaukee, WI, www.esca-tech.com (Last accessed 14 Aug. 2010).

[29] Mallinkrodt Baker Inc., *Material Safety Data Sheet (MSDS): EDTA Disodium Salt*, Mallinkrodt Baker, Phillipsburg, NJ, 2009.

[30] Frankild, S., Andersen, K., and Nielsen, G., "Effect of Sodium Laurel Sulfate on In Vitro Percutaneous Absorption of Water, Hydrocortisone and Nickel," *Contact Dermatitis*, Vol. 32, 1995, pp. 338–345.

[31] Esswein, E. J., Boeniger, M. F., and Ashley, K., "Wipes and Methods for Removal of Metal Contamination from Surfaces," US Patent No. 7,604,997 (2009).

[32] Klotz, A., Veeger, M., and Röcher, W., "Skin Cleansers for Occupational Use: Testing the Skin Compatibility of Different Formulations," *Int. Arch. Occup. Environ. Health*, Vol. 76, 2003, pp. 367–373.

[33] 2003, *ASTM E 1792, "Standard Specification for Wipe Sampling Materials for Lead in Surface Dust*, ASTM International, West Conshohocken, PA.

[34] U.S. National Institute for Occupational Safety and Health (NIOSH), "Development and Evaluation of a Novel Skin Cleansing System for Removing Lead and Other Toxic Inorganics," HSRB 04-DSHEFS-01XP, CDC/NIOSH, Cincinnati, OH, 2004.

[35] U.S. National Institute for Occupational Safety and Health (NIOSH), "Lead in Dust Wipes by Chemical Spot Test Method, (Colorimetric Screening Method) 9105," *NIOSH Manual of Analytical Methods*, CDC/NIOSH, Cincinnati, OH, 2003, www.cdc.gov/niosh/nmam (Last accessed 20 Sept. 2010).

[36] ASTM E1613-04, 2004, "Standard Test Method for Determination of Lead by Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-AES), Flame Atomic Absorption Spectrometry (FAAS), or Graphite Furnace Atomic Absorption Spectrometry (GFAAS) Techniques," Annual Book of ASTM Standards, Vol. 04.11, ASTM International, West Conshohocken, PA.

[37] Boeniger, M., "A Comparison of Surface Wipe Media for Sampling Lead on Hands," *J. Occup. Environ. Hyg.*, Vol. 3, 2006, pp. 428–434.

LEAD

Reprinted from JAI, Vol. 8, No. 3
doi:10.1520/JAI103466
Available online at www.astm.org/JAI

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Development of Two Sample Preparation Methods for Determination of Lead in Composite Dust Wipe Samples

ABSTRACT: Sample preparation methods for four-wipe composite dust wipe samples, used by nine different laboratories accredited under the U.S. Environmental Protection Agency (EPA) National Lead Laboratory Accreditation Program (NLLAP), were evaluated to select two methods most likely to provide the best overall performance among multiple laboratories, based on accuracy, precision and practicality of the methods. The best performing methods involved (1) hot nitric acid/hydrogen peroxide digestion and (2) nitric acid ultrasonication. In a second round of testing, two sets of composite dust wipe samples with known amounts of lead were submitted blind to each of the nine laboratories to be prepared for analysis using these two sample preparation methods. Results showed that both methods are capable of meeting all EPA NLLAP requirements for accuracy (recovery) and precision. The ultrasonication method had superior performance at lower cost.

KEYWORDS: composite wipes, lead analysis, lead-based paint

Introduction

Wipe samples of settled dust are used to assess the dust-lead hazard level on horizontal surfaces such as floors, window troughs, and interior windowsills in residences. Typically, each wipe sample of settled dust is collected from a single

Manuscript received October 14, 2010; accepted for publication January 26, 2011; published online March 2011.

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Cite as: White, K. T., Dewalt, F. G., Cox, D. C., Schmehl, R., Friedman, W. and Pinzer, E. A., "Development of Two Sample Preparation Methods for Determination of Lead in Composite Dust Wipe Samples," *J. ASTM Intl.*, Vol. 8, No. 3. doi:10.1520/JAI103466.

square foot of the floor being tested, or from the entire surface of an interior windowsill or window trough. Composite wipe samples result when up to four individual wipe samples collected from a single surface (e.g., a bedroom floor) are combined into a single sample. Regulations [1,2] and federal guidelines [3] allow use of composite samples for conduct of lead-based paint risk assessments, lead hazard screens, and post-abatement and post-interim controls clearance. Regulation also requires that all samples collected during conduct of these lead-based paint activities shall be analyzed by laboratories recognized by the U.S. Environmental Protection Agency (EPA) through the National Lead Laboratory Accreditation Program (NLLAP). Presently, there are no published methods for the preparation of composite wipe samples for subsequent analysis of lead content. This work was performed to establish acceptable methods for processing composite wipe samples and in anticipation of inclusion of composite samples in the Environmental Lead Proficiency Analytical Testing (ELPAT) Program [4] toward accreditation under the NLLAP.

Materials and Methods

General Approach

A five-step approach using two rounds of analyses was used to identify two acceptable methods for preparing composite wipe samples for lead determinations.

Step 1—Identify Existing Sample Preparation Methods —Nine laboratories, each quoting a different wipe sample preparation method, were chosen from the approximately 140 laboratories recognized by EPA through NLLAP as capable of analysis for lead in settled dust wipe samples. A summary of the sample preparation methods used by the laboratories for processing a four-wipe composite sample is provided in Table 1.

Step 2—Evaluate Existing Sample Preparation Methods (Round 1 Testing)—Each of the nine participating laboratories was instructed to use their own method (Table 1) to process a set of reference four-wipe composite wipe samples in a first round of testing.

Step 3—Select 2 Candidate Sample Preparation Methods—The results from round 1 testing were reviewed and analyzed to determine the two best performing candidate methods using three criteria: Accuracy, precision, and practicality of use.

Step 4—Evaluate Candidate Sample Preparation Methods (Round 2 Testing)—The same nine laboratories participating in round 1 testing were asked to participate in round 2 testing using the two candidate methods: The hotplate method (originating from Laboratory E from round 1) and the sonication method (originating from Laboratory B from round 1). Laboratory F withdrew citing that continued participation would interfere with their normal

TABLE 1—*Summary of sample preparation methods used during round 1 testing.*

Laboratory ID	Sample Preparation Summary	Instrumental Analysis
A	Transfer sample to 50 mL beaker w/watch glass cover, 1 mL con perchloric acid and 3 mL con nitric acid on hotplate until white fumes appear; rinse watch glass bottom and beaker side walls with water and add 1 mL con nitric acid; transfer/decant to 25 mL volumetric flask; add water to volume.	Inductively coupled plasma
B	Transfer sample to 125 mL screwtop bottle with top, add 15 mL con nitric acid; sonicate for 30 min; add water to make to 50 mL total liquid volume; sonicate for 30 min.	Flame atomic absorption
C	Split the sample into two sub-samples with two wipes each; place each sub-sample in 125 mL Erlenmeyer flask with watch glass cover, add 15 mL con nitric acid and 1 mL 30 % hydrogen peroxide on hotplate, reflux for 20 min; add water to 100 mL total volume.	Flame atomic absorption
D	Transfer sample to 250 mL or 500 mL beaker, add 25 mL con nitric acid, 20 mL water, and 6 mL con hydrochloric acid; hold the wipes below the liquid surface using a glass rod; heat in a boiling water bath for about 1 h; collect digestate by squeezing the wipes; rinse; add water to 50 mL volume.	Flame atomic absorption
E	Transfer sample to 250 mL beaker with watch glass cover, add 60 mL con nitric; heat on hotplate and reduce to 8 mL; cool; add 40 mL 30 % hydrogen peroxide; heat on hotplate and reduce to 8 mL; add water and filter into a 400 mL volumetric flask; rinse; add water to volume	Inductively coupled plasma
F	Split the sample into four sub-samples with one wipe each; place each sub-sample into the 67 mL hotblock container; heat to 100°C in hotblock; add 15 mL con nitric acid; heat for 1 h; add 5 mL con hydrochloric acid; heat for 5 min more; allow to cool; add 5 mL 30 % hydrogen peroxide; filter into 50 mL centrifuge tube; add water to 50 mL volume.	Inductively coupled plasma
G	Transfer sample to 150 mL beaker with watch glass cover; add 10 mL con nitric acid; heat on hotplate until the solution boils and continue boiling for 2 to 4 min; add 1 to 2 mL 30 % hydrogen peroxide aliquots until no color change seen; add con nitric acid to maintain liquid level; cool; filter into a 50 mL volumetric flask; rinse; add water to volume.	Flame atomic absorption
H	Transfer sample to 200 mL or 250 mL beaker with watch glass cover; add 20 mL 1:1 nitric acid; heat on hotplate at about 90°C for 2 h; replenish with 1:1 nitric acid to maintain volume; near the end add 5 mL 1:1 hydrochloric acid; filter into a 100 mL volumetric flask; add water to volume.	Inductively coupled plasma
J	Transfer sample to 250 mL beaker with watch glass cover; add 20 mL con nitric acid; heat on hotplate to boil until no fumes visible; allow to cool; add 3 mL 30 % hydrogen peroxide and boil for 10 min; allow to cool; add 5 mL con nitric acid and boil for 10–15 min; allow to cool; filter or centrifuge; transfer to 100 or 150 mL volumetric flask; add water to volume.	Flame atomic absorption

laboratory operations. The remaining eight participating laboratories were provided with the two written test methods and were sent two sets of reference four-wipe composite wipe samples (one for processing by each method).

Step 5—Develop Guidance on the Suitability of the Candidate Sample Preparation Methods—The results from round 2 testing were reviewed and analyzed to provide guidance on sample preparation procedures for laboratory processing of four-wipe composite dust wipe samples for lead analysis.

Round 1 Testing Samples

Each set of samples contained five reference samples, each in triplicate, and one blank as shown in Table 2. Each reference sample contained four wipes. In each sample, the sample reference material is present on one wipe and three additional blank wipes were added to the sample containers to achieve the four-wipe composite. The lead content of all samples was unknown to the laboratories (single blinded).

Round 1 Testing Analysis—Accuracy and Precision

Accuracy and precision were determined by performing a one-way analysis of variance for each laboratory using percent recovery as the dependent variable and the five certified lead levels as treatments. Mean percent recoveries were used to measure accuracy and the pooled estimate for the standard deviation was used to measure precision.

Round 1 Testing Analysis—Practicality of Use

Practicality of use among methods showing acceptable recovery from round 1 testing was evaluated using the operational parameters shown below. The instrumental analysis methods used by all the laboratories, shown in Table 1, were considered equivalent and were not included in this evaluation.

TABLE 2—*Sample set summary for round 1 testing (all samples contained four individual wipes).*

Sample Type	Lead Loading [μg/sample]	Number of Samples in Test Set
ELPAT[a] 23, w3	59	3
ELPAT 19, w4	127.1	3
ELPAT 23, w2	271.5	3
ELPAT 23, w4	562	3
NIST SRM[b] 2582 (0.2400 g)	50	3
Blank	0	1

[a]AIHA ELPAT Program. Sample round number and identifiers are shown.
[b]NIST SRM.

(1) *Relative complexity*. This parameter refers to the overall degree of complexity of the method. Methods with a large number of steps or multiple transfers of sample material would receive a higher complexity rating than those with less steps and easy manipulations.
(2) *Specialized equipment use*. This parameter refers to the overall need for specialized equipment that may not be commonly present in laboratories, especially smaller ones. Use of specialized equipment that is expensive (such as a perchloric acid hood) would rank higher than low-cost equipment (such as a sonication bath).
(3) *Use of uncommon or more hazardous reagents*. This parameter refers to the use of less common reagents. Use of uncommon or comparatively more hazardous reagents (such as perchloric acid) would rank higher than more common reagents (such as nitric acid).
(4) *Relative duration*. This parameter refers to the time required to process a sample or batch of samples. Longer duration methods would rank higher than shorter duration methods.
(5) *Relative labor use*. This parameter refers to the expected labor required to process a sample or batch of samples. Increased use of labor or use of more highly skilled labor would rank higher than less or lower skill level labor efforts.

Round 2 Testing Samples

Nine reference samples [eight ELPAT samples at various levels and one National Institute of Standards and Technology (NIST) Standard Reference Material (SRM)] were used in round 2 testing. However, only five ELPAT samples per level were available. These were equitably distributed between the two methods as alternating duplicates and triplicates as shown in Table 3. Each

TABLE 3—*Sample set summary for round 2 testing (all samples contained four individual wipes).*

Sample Type	Lead Loading [μg/sample]	Number of Samples in Test Set for Sonication Method	Number of Samples in Test Set for Hotplate Method
ELPAT[a] 17, w4	29	3	2
ELPAT 23, w3	59	2	3
ELPAT 19, w1	80.2	3	2
ELPAT 19, w4	127.1	2	3
ELPAT 23, w2	271.5	3	2
ELPAT 19, w3	287.1	2	3
ELPAT 23, w4	562	3	2
ELPAT 19, w2	1498.5	2	3
NIST SRM[b] 2582 (0.2400 g)	50	2	2
Blank	0	1	1

[a]AIHA ELPAT Program.
[b]NIST SRM.

reference sample contained four wipes. In each sample, the sample reference material was present on one wipe and three additional blank wipes were added to the sample containers to achieve the four-wipe composite. The lead content of all samples was unknown to the laboratories (single blinded).

Participating laboratories also received approximately 25 (14 plus spares) four-wipe composite wipe sample blanks with instructions to include seven of these blanks with the processing of each sample set after inoculating each of them with 30 μg of lead. These "spiked" composite wipe sample blanks were included for use in determining a method detection limit (MDL).

Round 2 Testing Analysis—MDLs

The NLLAP Laboratory Quality System Requirements (LQSR) [5] state that recognized laboratories shall determine the MDL at least annually and estimate the method quantification limit (MQL) to be at least two times but no greater than 10 times the MDL. By agreement, EPA and the NLLAP laboratory accrediting bodies have established that: The MQL for wipe samples of settled dust shall be equal to or less than one-half (50 %) of the Lowest Lead Level of Concern; and, the Lowest Lead Level of Concern for single wipe samples is 40 $\mu g/ft^2$, which is equal to the clearance levels established by EPA regulations at 40 CFR 745 [1]. Therefore, assuming a typical one square foot sampling area for a single wipe sample, the NLLAP LQSR required MQL for a single wipe sample is 20 μg and the required MDL is 10 μg.

The NLLAP does not directly address composite wipe samples. However, the EPA regulations at 40 CFR 745 [1], have established a clearance standard for floors, utilizing composite wipe sampling, as 40 $\mu g/ft^2$ divided by one-half the number of sub-samples in the composite sample, i.e., 20 $\mu g/ft^2$ for a four-wipe composite. Assuming a typical one square foot sampling area for each wipe, 20 $\mu g/ft^2$ equates to 80 μg of lead on the four-wipe composite. Therefore, the NLLAP LQSR implied MQL and MDL for a four-wipe composite sample are 40 and 20 μg, respectively [5].

MDLs were calculated using the lead results reported from seven spiked composite four-wipe sample blanks and the procedure described in 40 CFR Part 136 [6].

Round 2 Testing Analysis—Accuracy

The NLLAP LQSR requires that Laboratory Control Standards (LCS) be processed at a minimum rate of 5 % and that the recovery from these samples be within $\pm$20 % of the actual value. Laboratories frequently set the lead level of the LCS at or near the regulatory action level, which for a four-wipe composite collected from 4 ft^2 of a floor is 80 μg/sample (one-half of 40 $\times$ 4). In addition, the NLLAP LQSR directs that $\pm$20 % accuracy be demonstrated on a daily basis at the "Reporting Limit," which these authors interpret as equal to the allowable maximum MQL for a four-wipe composite, i.e., 40 μg/sample. Most of the samples in the sample sets distributed to participant laboratories (Tables 2 and 3) were assembled using samples from past rounds of the American Industrial

Hygiene Association (AIHA) ELPAT Program which is specifically named in the NLLAP LQSR as a source of LCSs. Therefore, the results from round 2 testing should be predictive of likely method recoveries from accredited laboratories using the two methods. The mean recoveries on the reference samples processed by each laboratory for each method were examined using the NLLAP LQSR criterion of ±20 % as acceptable performance.

In addition, a statistical evaluation of accuracy at the 40 and 80 μg levels was performed by generating percent recovery curves as a function of lead level for each laboratory using each method. These curves were estimated as shown in Table 10 below using the following power equation:

$$\text{Percent recovery} = \alpha^{*}(\text{True lead level})^{\beta}$$

Curve fitting was accomplished by linear regression after taking logarithms of both sides of the equation.

Since analytical measurement variability was larger at lower analyte levels than at higher levels, precision estimates on the percent recovery were calculated using two roughly equal sized groups of reference samples: Those above and those below 160 μg. Precision was relatively constant within each of these groups for both candidate methods used in round 2 testing. The precision estimates were calculated by pooling variances of percent recovery within each group.

Results

Round 1 Testing

Sixteen lead results were obtained from each of the nine participating laboratories on its preferred method (15 reference sample results, and one blank sample result). The results of the one-way analysis of variance analyses, sorted by the mean percent recoveries, are shown in Table 4. The first column of Table 4 shows a code identifying the nine participating laboratories as A through J (the letter I is not used). The second column shows the mean percent recovery from each laboratory for the 15 samples analyzed. The third column shows the pooled estimate of the standard deviation of percent recovery of a single measurement for each laboratory across all lead levels. The fourth column shows the *p*-value for statistically testing the validity of pooling the recoveries from all five lead levels in the reference sample sets to create variance estimates for a single measurement. Pooling of the recoveries from all five lead levels to calculate an estimated standard deviation is only valid if the true variability across all of the lead levels is the same. Using a 95 % confidence interval, *p*-values less than 0.05, as shown for Laboratories D and H, suggest differences in variability as a function of lead level.

Practicality of use results among the laboratories having mean percent recovery for round 1 testing of between 95 and 105 % are shown in Table 5 (Laboratory C was excluded because of a high standard deviation). The first column

TABLE 4—*Summary of analysis of variance results for round 1.*

Laboratory Code	Mean Percent Recovery	Pooled Estimate of Standard Deviation (Using 10 Degrees of Freedom)	*p*-value
D	76.6	4.17[a]	0.02
G	80.8	5.42	0.20
J	90.5	7.63	0.29
H	94.2	6.37[a]	0.01
B	95.2	6.55	0.39
F	95.4	8.88	0.19
C	99.8	19.54	0.54
E	101.4	8.92	0.33
A	104.1	3.84	0.31

[a]The *p*-values suggest that pooling of variability estimates is not valid for these laboratories.

shows the evaluated parameter. The next four columns show the relative ranking for the parameter among the four laboratories (A, B, E, and F). A three-level ranking (low, moderate, or high) was used for this evaluation, which was performed by a pair of senior level chemists with experience in laboratory operations for metals analysis. Comments relevant to the ranking made by the chemist are provided in the last column. The method used by Laboratory B has the lowest overall ranking (rated best) followed by Laboratory E with Laboratories F and A having one or more "high" (or poor) rankings. Therefore, the methods used by Laboratories B and E were selected for round 2 testing.

Round 2 Testing

Thirty lead results were obtained from seven of the eight participating laboratories on each of the two candidate methods (22 reference sample results, one blank sample result and seven spiked blank sample results). Laboratory A failed to use the hotplate method and instead processed both sets of (22 + blank =)23 samples using only the sonication method generating 46 lead results from the two reference sample sets and seven lead results on spiked composite wipe sample blanks. Laboratory A lead results from the 23 samples originally targeted for the hotplate method were excluded from data analysis. In addition, two other lead results from Laboratory G were also excluded as statistical outliers. The first outlier was a 252.4 μg lead result reported using the sonication method on a 1498.5 μg ELPAT sample. Using the sonication method for the replicate ELPAT sample present in that same sample set reported a 1570 μg lead result. The second outlier was a zero lead result reported using the hotplate method on a 29 μg ELPAT sample. Using the hotplate method for the replicate ELPAT sample present in that same sample set reported a 20.59 μg lead result.

TABLE 5—*Summary of practicality of use considerations.*

Parameter	Laboratory ID				Comments
	A	B	E	F	
Relative complexity	Moderate	Low	Moderate	High	F: Four-way splitting of samples
Specialized equipment use	High	Moderate	Low	Moderate	A: Perchloric acid hood B: Sonication bath F: Hotblock
Uncommon reagents use	High	Low	Moderate	Moderate	A: Perchloric acid
Relative duration	Moderate	Low	Moderate	Moderate	
Relative labor use	High	Low	Moderate	Moderate	

TABLE 6—*MDLs from round 2 testing.*

Laboratory	Sonication MDL[μg/sample]	Hotplate MDL[μg/sample]
A	1.61	na[a]
B	4.12	17.99
C	10.60	35.00
D	4.80	6.70
E	6.72	24.00
G	7.37	34.92
H	2.31	4.88
J	2.49	16.39

[a]Not available. Laboratory A did not report any results for the hotplate method.

MDLs for the participating laboratories are shown in Table 6. The first column shows a code identifying the participating laboratory. The second and third columns show calculated MDL for the hotplate and sonication methods, respectively. Four out of seven laboratories were able to achieve the NLLAP LQSR implied MDL criterion of 20 μg using the hotplate method while all of the laboratories were able to achieve this requirement using the sonication method.

Table 7 presents pooled estimates of the precision (percent recovery) for the two ranges of lead levels. The first column shows a code identifying the participating laboratory. The second and third columns show pooled estimates of the standard deviation of the percent recoveries for levels below and above 160 μg, respectively. The percentages in Table 7 can be interpreted as the precision (one standard deviation) of a single measurement as a percent of the true lead level. For example, the precision of a measurement at 40 μg of lead using the

TABLE 7—*Pooled estimates of the standard deviation of percent recovery.*

	Lead Levels Below[a] 160 μg		Lead Levels Above[b] 160 μg	
Laboratory	Sonication(%)	Hotplate(%)	Sonication(%)	Hotplate(%)
A	7	N/A	3	N/A
B	9	22	3	20
C	10	12	5	7
D	7	17	6	3
E	19	10	13	2
G	13	41	3	10
H	6	16	3	7
J	8	34	6	4

[a]Calculated using seven degrees of freedom: 12 reference samples minus five different lead levels except for Laboratory G using hotplate where one statistical outlier was removed resulting in six degrees of freedom.

[b]Calculated using six degrees of freedom: 10 reference samples minus four different lead levels except for Laboratory G using sonication where one statistical outlier was removed resulting in five degrees of freedom.

TABLE 8—*Recoveries for the sonication method for low and high lead level ranges.*

	Lead Levels Below 160 μg			Lead Levels Above 160 μg		
Laboratory	< 80 %	80–120 %	> 120 %	< 80 %	80–120 %	> 120 %
A	1	11	0	0	10	0
B	1	10	1	0	10	0
C	7	5	0	0	10	0
D	2	10	0	0	10	0
E	2	5	5	1	8	1
G	6	6	0	1	9	0
H	1	11	0	0	10	0
J	5	7	0	0	10	0
Total	25	65	6	2	77	1

sonication method in laboratory D is ±7 % or ±2.8 μg at one standard deviation. Thus, on average, 95 % of measurements will fall within ±14 % or ±5.6 μg of 40 μg (two standard deviations).

Tables 8 and 9 summarize the number of samples having recoveries in various ranges for the sonication and hotplate methods, respectively, for samples with lead levels above or below 160 μg. For the sonication method, 142 of the 176 total results or 81 % of the recoveries met the NLLAP LQSR criterion for recovery (80–120 %). For the hot-plate method, 114 of the 154 total results or 74 % of the recoveries met the NLLAP LQSR criterion.

Table 10 provides curve fit parameters for the percent recovery curves as a function of lead level for each laboratory using each method. The first column shows a code identifying the participating laboratory. The next four columns show the parameters for the sonication and hotplate method, respectively. For the sonication method, all but one of the β exponents in the power curve are positive, indicating typically increasing recovery with increasing lead level. All

TABLE 9—*Recoveries for the hot-plate method for low and high lead level ranges.*

	Lead Levels Below 160 μg			Lead Levels Above 160 μg		
Laboratory	< 80 %	80–120 %	> 120 %	< 80 %	80–120 %	> 120 %
A	N/A	N/A	N/A	N/A	N/A	N/A
B	0	7	5	2	8	0
C	0	2	10	3	7	0
D	0	10	2	0	10	0
E	0	11	1	0	10	0
G	5	3	4	1	9	0
H	0	10	2	0	10	0
J	3	7	2	0	10	0
Total	8	50	26	6	64	0

TABLE 10—*Curve fits parameters for percent recovery for round 2.*

	Sonication Method		Hotplate Method	
Laboratory	α	β	α	β
A	81	0.02[a]	N/A	N/A
B	114	−0.04	212	−0.13
C	52	0.10	372	−0.19
D	70	0.06	121	−0.03[a]
E	95	0.02[a]	101	−0.01[a]
G	45	0.12	72	0.05
H	77	0.04	125	−0.03[a]
J	57	0.09	82	0.03[a]

[a]Coefficient not statistically significant.

but two of the power coefficients for the hotplate method are negative so that recovery is typically decreasing with increasing lead level. Figures 1 and 2 show the fitted curves up to lead levels of 200 μg/sample by laboratory for the sonication and hotplate methods, respectively. The figures demonstrate graphically the inter-laboratory differences in recovery. Above 200 μg all plots stay within the ±20 % recovery criterion, with the exception of laboratory C for the hotplate method, which reaches the ±20 % range at a level of approximately 450 μg.

Table 11 provides an estimate of the percentage of samples achieving a recovery between the LQSR limits of 80 to 120 % at two lead levels for each

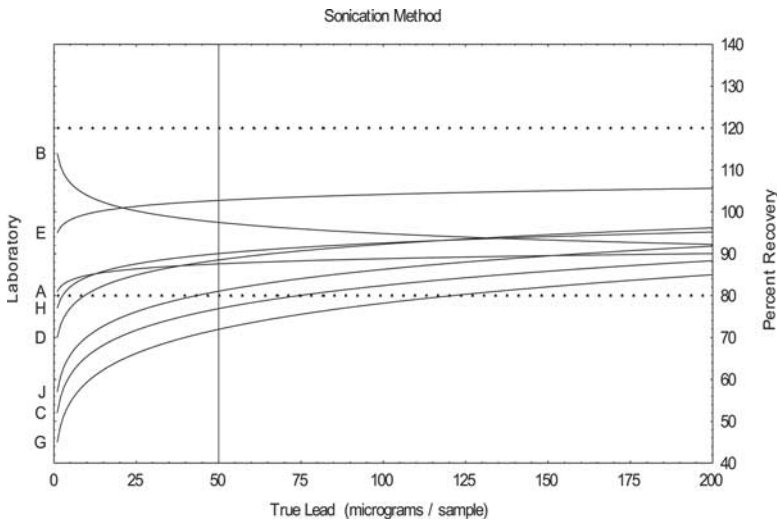

FIG. 1—*Fitted curves of recovery for the sonication method.*

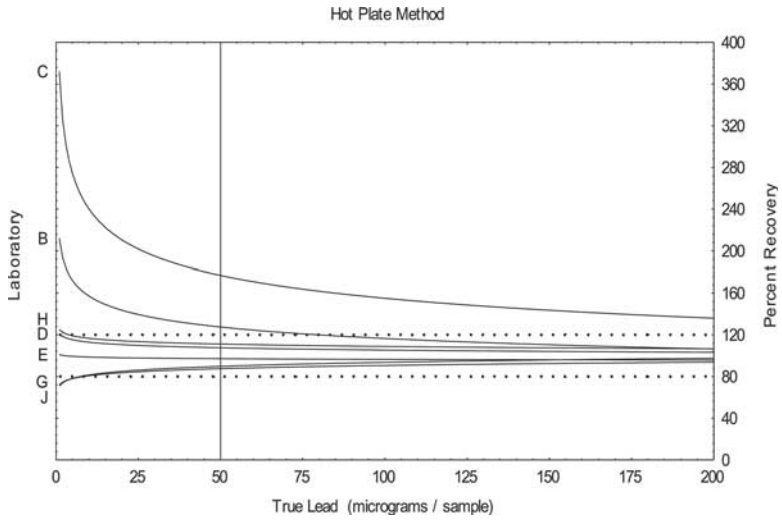

FIG. 2—*Fitted curves of recovery for the hotplate method.*

method: The NLLAP-LQSR-implied MQL of 40 μg, and, assuming a typical one square foot sampling area for each of four wipes, the federal Action Level would be 80 μg. The percentages of samples whose recoveries would fall between the 80 and 120 % limits at 40 and 80 μg lead, respectively, for each laboratory and method, as shown in Table 11, depend on both the overall accuracy as estimated by the power curve (Table 10) and the precision (Table 7). The percentages in Table 11 were calculated assuming that, for each laboratory and method, the recovery has a normal distribution with *mean* given by the power curve (at 40 and 80 μg, respectively) and *standard deviation* given in Table 7 for samples with less than 160 μg lead. The first column of Table 11 shows a code identifying the

TABLE 11—*Estimated percentage of samples with recoveries between 80–120 % by method and laboratory for two lead levels (40 and* 80 μg*).*

	Sonication Method		Hotplate Method	
Laboratory	40 μg(%)	80 μg(%)	40 μg(%)	80 μg(%)
A	86	**90**	N/A	N/A
B	**97**	**95**	31	48
C	32	52	0	0
D	82	**93**	69	72
E	69	69	**95**	**95**
G	26	43	35	36
H	**91**	**96**	67	71
J	53	77	43	43

participating laboratory. The next four columns show the calculated percentage of samples that would achieve the acceptable recovery limits at the two lead levels for the sonication and hotplate method, respectively. All percentages 90 % or greater are in bold.

Discussion

Selection of Candidate Methods from Round 1 Testing

Using the recovery data in Table 4, the methods used by Laboratories D, G, J, and H were removed from further consideration as candidate methods for round 2 testing. The method used by Laboratory C was also removed from further consideration because its estimated standard deviation is more than twice as high as the next closest method (Laboratory E). Of the remaining methods from Laboratories B, F, E, and A, the method from Laboratory E has the best mean recovery and the methods from Laboratories B and E were rated most practical (Table 5). Therefore, the methods from Laboratory B and E were selected as candidates for round 2 testing.

Performance of the Candidate Methods from Round 2 Testing

MDLs—Although both methods appeared to be capable of achieving the NLLAP LQSR implied MDL as shown in Table 6, the overall MDL performance of the sonication method was superior to the hotplate method in that all eight laboratories achieved MDLs below 20 μg for this method while only four of seven did so for the hotplate method.

Precision—The data in Table 7 indicates that both methods have roughly equivalent precision for higher lead levels, but precision of the sonication method for the lower lead levels is superior to that of the hot-plate method. At the lower lead levels, six of eight laboratories have precision of 10 % or less (one standard deviation) for the sonication method, while only one of seven does for the hotplate method.

Accuracy—The data in Tables 8 and 9 indicate that across all the participating laboratories, both methods were capable of meeting the NLLAP LQSR criterion of $\pm$20 % for most ($>$ 70 %) of the samples. However, these data also suggest performance differences between laboratories with both methods.

Using data from Table 11 for the sonication method, four laboratories (A, B, D, and H) have 90 % or more of recoveries between 80 and 120 % at the Action Level of 80 μg; two laboratories (B and H) do this well at the Reporting Limit of 40 μg. For the hot-plate method, only laboratory E has 90 % or more of recoveries between 80 and 120 % at either level.

It is interesting to note, using the curve fits (Table 10) shown graphically in Fig. 1, that the laboratory with prior experience using the sonication method, Laboratory B, has a very different response curve from the others showing decreasing recovery with increasing lead level. The reason for this is unknown.

A systematic positive laboratory lead contamination problem could account for this effect. However, no such indication was observed given that all zero lead level reference samples in round 2 generated results below the calculated MDLs for each respective laboratory. The laboratory with prior experience using the hotplate method, Laboratory E, shows an expected outcome from having experience with a method; a relatively constant recovery close to 100 % across the entire lead level range.

Conclusions

The study demonstrated that both methods are capable of meeting NLLAP LQSR requirements for MDLs, MQLs, and recovery for four-wipe composites. However, the sonication method proved superior in all areas, having lower individual laboratory MDLs and superior accuracy and precision as compared to the hot-plate method. It should, of course, be remembered that this was the first application of either method to four-wipe composites [except possibly for laboratory B (sonication) and laboratory E (hot plate)]. Performance on both methods would improve with practice and it is possible that the observed superiority of the sonication method would be reduced or eliminated over time. Nevertheless, the participating laboratories commented that the sonication method is the more favorable method when attributes like cost of reagents, technician time, and labware expense are considered. They reported that compared to the hotplate procedure, the sonication procedure:

- Requires a smaller amount of acid;
- Requires less technician time for conduct of the procedure (e.g., one laboratory reported that it required two full person days to hotplate-digest the sample set); and,
- Requires no glass labware (i.e., no dishwashing required of the 400 mL beakers and large volumetric flasks used with hotplate digestion) compared with the hotplate method.

Both methods have been submitted as draft standards to ASTM International Subcommittee E06.23 on Lead Hazards Associated with Buildings.

Acknowledgments

Financial support from the U.S. Department of Housing and Urban Development Office of Healthy Homes and Lead Hazard Control under Contract No. C-OPC-21358 is gratefully acknowledged. The writers also express appreciation to Charles Bond, Eric Botnick, Shamsher Brar, Mark Crain, Norma Roadcap, Dave Sandusky, Karen Tinklenberg, Joe Unangst, and Howard Varner for assistance during the conduct of this work.

References

[1] "Lead-Based Paint Poisoning Prevention in Certain Residential Structures," Title 40, Code of Federal Regulations, Part 745.

[2] "Lead-Based Paint Poisoning Prevention in Certain Residential Structures," Title 24, Code of Federal Regulations, Part 35.
[3] U.S. Department of Housing and Urban Development, 1995, "Guidelines for the Evaluation and Control of Lead-Based Paint Hazards in Housing," Washington, D.C., June 1995 as amended.
[4] American Industrial Hygiene Association (AIHA) Proficiency Analytical Testing Programs (PATP), LLC, 2700 Prosperity Avenue, Suite 250, Fairfax, VA 22031.
[5] U.S. Environmental Protection Agency, 2007, "EPA National Lead Laboratory Accreditation Program Laboratory Quality System Requirements," Rev. 3.0, Washington, D.C. (July 5).
[6] "Guidelines Establishing Test Procedures for the Analysis of Pollutants, Appendix B—Definition and Procedure for the Determination of the Method Detection Limit—Revision 1.11," Title 40, Code of Federal Regulations, Part 136.

Reprinted from JAI, Vol. 8, No. 8
doi:10.1520/JAI103469
Available online at www.astm.org/JAI

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Improving the Confidence Level in Lead Clearance Examination Results through Modifications to Dust Sampling Protocols

ABSTRACT: Multiple pre-cleaned, four-room residential containments were each subjected to a single high-dust generating activity involving lead-based paint. After dust generation, a geometric wipe sampling grid was used, with a target of 8 interior and 8 perimeter samples in each room, to assess dust-lead on the floors. The containment was then cleaned using the "High Efficiency Particulate Air (HEPA) vacuum/3-bucket wet-mop/HEPA vacuum" procedure found in the U.S. Dept. of Housing and Urban Development Guidelines. Post-cleaning (clearance) floor wipe samples were then taken at side-by-side locations to the pre-cleaning samples. It was found that: [1] floor dust-lead along the perimeters of rooms was three times more difficult to clean than dust-lead from the interiors of the rooms; [2] post-cleaning dust-lead loadings tended to be higher along the perimeters of the rooms than in the interiors of the rooms such that clearance failure was much more likely for individual floor samples collected along the perimeters; and [3] four-wipe composite sampling within each room (two randomly selected from the perimeter and two randomly selected from the interior) provided a very reliable method for detecting clearance failure (99% or greater) versus a randomly selected single wipe sample per room (50% or less).

Manuscript received October 14, 2010; accepted for publication June 16, 2011; published online August 2011.

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Cite as: Cox, D. C., Dewalt, F. G., White, K. T., Schmehl, R., Friedman, W. and Pinzer, E. A., "Improving the Confidence Level in Lead Clearance Examination Results through Modifications to Dust Sampling Protocols," *J. ASTM Intl.*, Vol. 8, No. 8. doi:10.1520/JAI103469.

KEYWORDS: clearance testing, lead analysis, lead-based paint, composite wipes

Introduction

The most common pathway of childhood exposure to lead is through the ingestion of leaded dust. In response, a key component of the U.S. Dept. of Housing and Urban Development (HUD) Lead Safe Housing Rule (24 CFR Part 35) [1] is the required use of clearance testing as a check on the effectiveness of cleaning following a dust-generating activity. Clearance testing is aimed at ensuring that fine particles of lead in dust have been cleaned up before living spaces are reoccupied.

The conduct of clearance as defined by EPA protocols found at 40 CFR Part 745.227 [2] includes only minimal amounts of testing on floors. It includes the collection of "one floor sample from each of four rooms inside the containment area" and if containment exists, "one floor sample outside the containment area." No guidance is provided on where these floor samples should be collected. Because of the difficulties inherent in making changes to current regulations that already define the minimum number of samples to be collected for clearance, this study focused on constructive improvements within the regulatory framework, such as guidance on the sampling strategy. However, the impact of increasing the number of samples, not prohibited by the EPA protocols, was also examined. Improved guidance in order to better detect true clearance failures and protect occupants was a desired outcome of this work.

The greatest need for improved sampling guidance is when post-activity cleaning is inadequate and leaves dust-lead unequally distributed (i.e., sometimes above and sometimes below the federal action level for lead in dust) on the area to be cleared. Under a normal lead hazard reduction scenario, the environment goes from having dust-lead levels well above the action levels (i.e., just after conduct of the work and before the cleaning has begun) to levels well below the action levels after cleaning is performed to remove the dust. It is evident that, if the true dust-lead levels after cleaning are either all well below or all well above the action level throughout the work area, then it is not critical where in the work area dust samples are collected. It is in situations where the true residual dust-lead levels vary both above and below the action level in the same room that guidance on sampling is most likely to improve the ability to detect true clearance failures, and therefore protect against childhood lead poisoning.

Materials and Methods

The general study procedure was to prepare (clean) suitable rooms within a defined work area of a unit, disturb paint from a single point source in selected rooms within the work area, map the resulting dust lead levels (so-called post-activity samples), conduct standardized cleaning to remove the generated lead

hazards, and re-map the dust lead levels remaining after cleaning (so-called post-cleaning samples). Details on the field activity steps 1 through 8 are discussed below.

Activity 1-Select Housing Units

Suitable housing candidates for this study were located through the Mahoning County (Ohio) Healthy Homes and Lead Hazard Control Program for the Youngstown, OH units and through the Redevelopment Authority of the City of Erie for the Erie, PA units. Candidates for initial in-field examination were identified using a set of criteria designed to fit the technical and logistical needs of the study. In Youngstown, OH, four dwelling units (assigned unit numbers 2 through 5) were selected for this study out of 12 candidates. All of these dwellings were privately owned and scheduled to receive lead hazard reduction funding through the HUD lead hazard control grant program. In Erie, PA, four dwelling units (assigned unit numbers 6 though 9) were selected out of seven candidates. These dwellings were owned by the City of Erie.

All the selected units were structurally sound pre-1978 dwellings, unoccupied at the time of the study but with basic utilities available (water, electricity, and heat). Only hard floors (uncarpeted) were selected from each unit. Residents' belongings, if present, were removed. All floors were dust strewn and showed much wear. Some of rooms in the study had until recently been covered with wall-to-wall carpeting.

For this study, each dwelling unit was to have a minimum of three "active" rooms and up to two "passive" rooms if these additional rooms were adjacent to an active room and adjoined by an open door or passageway, making them part of the work area. Active rooms are rooms containing lead-based paint where a single point source of leaded paint was reduced to dust using mechanical grinding. Passive rooms are rooms where no leaded dust was generated. Passive rooms were included within the work area to evaluate the potential lead hazard impact caused by performing lead-disturbing activity in an adjacent room. All tested rooms in a given unit were located on a single floor.

Portable x-ray fluorescence (XRF) analysis was used both to screen unit candidates for inclusion and for selection of potential painted surfaces to be disturbed for leaded dust generation. In seven out of eight of the housing units used in the study, paint chips were also collected as a means for obtaining better values for the lead-based paint sources targeted for dust generation.

Activity 2-Phase A Sampling

This phase of sampling included the collection of pre-existing condition dust wipe samples in the rooms selected for inclusion in the work area. These were collected as a hedge against unusual findings that might be clarified by knowing what the dust lead levels were immediately preceding the field activities. All dust wipes used for sample collection in this study were individually packaged dust wipes conforming to ASTM E1792 [3].

Activity 3-Room Preparation and Cleaning

Barriers were erected as needed to isolate the set of rooms being used for the study from the rest of the unit and from the outside. This was done both as a means of limiting the potential spread of generated leaded dust to rooms outside the work area and to help reduce the impact from external air currents on movement of leaded dust during the conduct of the study. In most cases these barriers consisted of sheets of 2 mil (0.508 mm) poly fixed over open doorways and windows as needed to complete the isolation.

All horizontal surfaces in the selected rooms were pre-cleaned using a High Efficiency Particulate Air (HEPA) vacuum followed by a wet mop cleaning. A second HEPA vacuuming was done in the housing units in Erie but not in Youngstown due to in-field time constraints. This ensured removal of any pre-existing debris and dust that might contribute in any significant manner to the final lead loading resulting from dust generation.

Activity 4-Sample Location Selection and Marking

In each of the eight dwellings, 120 dust wipe floor samples were targeted for collection: sixty before the cleaning treatment and sixty after the cleaning treatment. The *before* and *after* samples were physically paired on the floor to allow for paired comparisons of the Before- and After-Cleaning treatments.

Samples were designated as either "wall" or "interior" samples. A wall sample was against a wall (or other vertical structure such as an installed cabinet or other vertical barrier, such as a door). An interior sample was one located away from the walls in the interior of the room or in a wall opening without a door. Samples were symmetrically arranged within each room to produce a lead-loading map of the floor, but the number of samples collected per room varied depending on the number and size of the rooms. A four-room unit typically had a target of sixteen samples for each active room (eight wall and eight interior samples) and twelve samples for the single passive room. The active room typically had two wall samples per wall and eight interior samples, arranged in rows of four samples each. In many cases; however, these sample allocations were not exactly attained, because of unusual room shapes, or multiple doors or other openings.

Sampling locations on the floors of each selected room were marked with the aid of an aluminum U-shaped and square-shaped templates with an internal area of 1 ft^2 (0.0929 m^2). For each location, a sample location number was marked on the floor in roughly the center of the template before sample collection. In addition, the four inside corners of the template were marked on the floor. Marking pens that resisted removal using normal cleaning processes were used. A pair of side-by-side locations was marked for each sampling location. One side of the pair was randomly selected for collecting the before-cleaning treatment samples and other for collecting the after-cleaning treatment samples.

Activity 5-Dust generation-Lead Loading Model

Dust was generated from a single source point in each of the active rooms by converting a defined small surface area of existing lead based paint into dust.

An important component of the study was to generate post-cleaning dust-lead levels that varied both above and below the action level (40 $\mu g/ft^2$ for floors) so that guidance on sampling that is most likely to improve the ability to detect true clearance failures could be determined. As a result, a procedure was developed using a pilot study to obtain after-cleaning lead levels that were neither too high nor too low to draw useful conclusions. The amount of leaded dust generated by disturbing a leaded paint surface is dependent on the activity that is used to disturb (break up) the paint. Limits in study resources did not allow for a detailed investigation of the ability of various work methods to generate leaded dust. Rather, for this study, mechanical grinding using a portable electric grinder was used to generate leaded dust. Although this method would be considered a forbidden dry paint removal method in a renovation or removal project conducted under the HUD Guidelines [4], it was the only method that could be universally applied to any painted surface in any selected housing unit to consistently and rapidly generate large amounts of leaded dust.

The pilot study, conducted in one house in Youngstown (assigned unit number 1 as opposed to 2 through 9 for the full study), used two levels of paint disturbance: one deemed high and one deemed low. Five rooms were tested at both high and low levels of paint disturbance and the total mass of lead (mg) in dust deposited on the floor was calculated using the XRF-determined lead loading on the painted surface to be disturbed and the area of paint disturbed. The variable PB, equal to mg lead in dust deposited per square foot of floor area, was calculated by dividing the total mass of lead deposited by the area of each room. The variable MEAN, equal to the average lead loading in $\mu g/ft^2$ for (post-cleaning) "clearance" samples taken in each room, was also calculated, and the regression equation

$$\mathrm{Ln(MEAN)} = 3.269 + (0.991)(\mathrm{Ln(PB)})$$

was estimated using the 10 data points from the pilot (5 rooms, 2 levels of disturbance per room). In order to target post-cleaning floor lead levels close to the EPA standard (action level) of 40 $\mu g/ft^2$ for floors, the regression equation was solved for PB with MEAN = 40. The result was that an estimated 1.53 mg of lead must be disturbed per square foot of floor area in the room in order to achieve a target level of 40 $\mu g/ft^2$ on the floor post cleaning. In the full study, comprised of the 4 units in Youngstown and 4 units in Erie, paint samples were taken in the targeted units and analyzed in the laboratory (as opposed to relying on XRF data) to improve the accuracy of predicted post-cleaning levels. The lab data was then used to calculate the area of paint to disturb in each room of each unit. For example, UNIT 2, ROOM 1 had a lead level of 2.9 mg/cm^2 by laboratory analysis, and a floor area of 220 ft^2. Thus, the target total paint area to be disturbed was estimated at $(1.53)(220/2.9) = 116\ cm^2 = 18\ in^2$ [2].

Table 1 summarizes the dust generation calculations for the full study. The actual areas disturbed were measured in the field and differ somewhat from the model calculations, for two reasons. First, the area disturbed cannot be precisely controlled in the field. Second, the model was adjusted slightly for the

TABLE 1—*Summary of Lead Dust Generation by Room.*

Unit ID	Room ID	Estimated Lead in Paint Disturbed (mg/cm^2)	Approximate Painted Area Disturbed (cm^2)	Approximate Area of Room (ft^2)	Calculated Lead Loading If Dust Spread Uniformly ($\mu g/ft^2$)
2	1	2.9	135	220	1814
2	2	1.8	135	140	1728
2	5	3.4	32	123	902
3	1	9.3	19	126	1430
3	2	11.2	29	139	2353
3	4	5.8	68	265	1489
4	2	1.5	157	147	1574
4	3	1.5	270	245	1666
4	4	13.2	19	166	1534
5	1	2.8	115	219	1464
5	2	1.4	139	123	1525
5	3	0.9	172	93	1573
6	1	6.2	69	159	2684
6	2	2.8	44	187	662
6	4	5.0	45	185	1229
7	1	0.7	181	100	1189
7	3	7.4	30	184	1213
7	5	5.4	43	193	1218
8	1	5.4	37	168	1211
8	3	6.6	31	148	1380
8	4	3.3	56	172	1088
9	1	5.8	21	134	925
9	3	3.4	60	174	1189
9	4	3.1	44	112	1195

second city in the full study (Erie, PA), based on the results for the first city (Youngstown, OH).

Activity 6-Phase B Sampling

The first of a pair of side-by-side dust wipe samples in each selected room was collected following dust generation using standard dust wipe collection procedures based on ASTM E1728 [5]. The sampling order was planned such that care was taken to avoid stepping on or disturbing the other locations to be sampled.

Activity 7-Cleaning Treatment

Cleaning was conducted using the widely-accepted HEPA vacuum/3-bucket-wet-clean/HEPA vacuum method as recommended in the HUD Guidelines [4].

Activity 8-Phase C Sampling

The second of the pair of side-by-side samples in each selected room was collected following dust generation and cleaning treatment using the same sampling procedure as Phase B sampling. Additional cleaning efforts were performed after this sampling to ensure that the units were safe for release back to the agencies providing the dwellings.

Data Processing, Laboratory Analysis and Quality Control

All dust wipe samples were assigned unique identification numbers (IDs) using pre-printed labels, and were traceable from field generation to laboratory processing using standard chain-of-custody procedures. Samples were shipped to a laboratory accredited under the National Lead Laboratory Accreditation Program (NLLAP) and analyzed for lead analysis by ultrasonic-assisted acid digestion based on ASTM E1979 [6], followed by Inductively Coupled Plasma Atomic Emission Spectrometry based on ASTM E1613 [7]. Data were reported both electronically and in hardcopy formats. A final analysis data set was created by merging electronic data received from the laboratory with data from field data collection forms that were manually keyed into the database.

In addition to standard quality control (QC) performed by the laboratory as part of their normal operations needed to maintain accreditation [8,9], two more types of QC samples were collected for this study: field blanks and QC reference wipes.

- The *field blank wipe* sample was a new lead-specific dust wipe exposed to the air in the study area before placing it in the labeled hard-shell container. The number of field blank wipes targeted for collection is summarized below:
  - Phase A. Two field blanks per unit: one before and one after collection of other samples.
  - Phase B. Four field blanks per unit: one before and one after collection of other samples. Two taken during collection of other samples.
  - Phase C. Four field blanks per unit: one before and one after collection of other samples. Two were collected during collection of other samples.
- The *QC reference wipe* samples, obtained from Research Triangle Institute, were used as double blind samples to validate laboratory performance. These samples were developed for use by the Environmental Lead Proficiency Analytical Testing Program (ELPAT) for the NLLAP [9]. The ELPAT samples used in this study ranged from 17 to 1499 μg lead per sample. These "old ELPAT" samples were placed in labeled hard-shell containers outside the study area in a lead-dust-free environment and inserted blindly into groups of samples sent to the laboratory for analysis. The number of QC reference samples included in each group is summarized below:
  - Phase A. Two QC reference samples per unit.
  - Phase B. Four QC reference samples per unit.
  - Phase C. Four QC reference samples per unit.

Six of the 79 field blanks collected for the study showed detectable levels of lead above the laboratory reporting limit of 5 μg (one field blank targeted for

collection was inadvertently not collected). Those levels ranged from 5.4 to 13.5 μg of lead. The majority of these (5 out of 6) occurred during the before-cleaning sampling (Phase B) when lead dust loadings were the highest. These results do not raise concerns about the handling of samples during collection, since they are only slightly above the reporting limit and are much lower than the pre-cleaning study samples.

Eight of the 80 QC reference samples showed recoveries outside the desired recovery range of 80% to 120%. All showed recovery within 70% to 133%. These results likewise do not raise concerns about the laboratory analytical process.

Results

Analysis results of pre- and post-cleaning floor samples make up a data set that covers 476 table rows (without headers) and includes X-Y coordinates designating the locations of each sample in each room of the 8 units in the study. Because of its large size, it is not included here. However, various summaries of this data are provided under the discussion section. To obtain a better understanding of the results, scale floor plan diagrams of all 33 study rooms were created showing the locations and results of pre- and post-cleaning floor samples ($\mu g/ft^2$). Examples of these diagrams are shown in Figs. 1 through 3. The post-cleaning or clearance results, shown below sample location IDs (those with "B" or "C" suffix codes), are in normal text font for samples with loadings below 40 $\mu g/ft^2$ (i.e., samples that pass clearance) and are in bold italics for samples with loadings of 40 $\mu g/ft^2$ or greater (i.e., samples that would fail clearance).

Discussion

Comparison of Interior Samples and Wall Samples

An examination of all 33 floor plan diagrams (three of these are shown in Figs. 1 through 3) show that, except in rooms where all or almost all post-cleaning samples fail clearance, samples taken against the wall are more likely to fail clearance than samples taken from the interior of the room or in openings without doors. Table 2 shows the sample counts and number of clearance failures by room and unit for INTERIOR and WALL samples. A sample is classified as INTERIOR if it is taken in the interior of the room or in an opening to the room with no door. A sample is classified as WALL if it is taken against a vertical surface in the room, such as wall, cabinet, mirror, etc., or in an entry to the room with a door. The rooms are classified as ACTIVE if the grinding process used to generate lead dust was actually carried out in the room. These rooms have a "SOURCE" location shown on the diagram as shown in Figs. 1 and 3. Other rooms, without a SOURCE, such as the room shown in Fig. 2, are termed PASSIVE. Such rooms were adjacent to active rooms where no barriers were

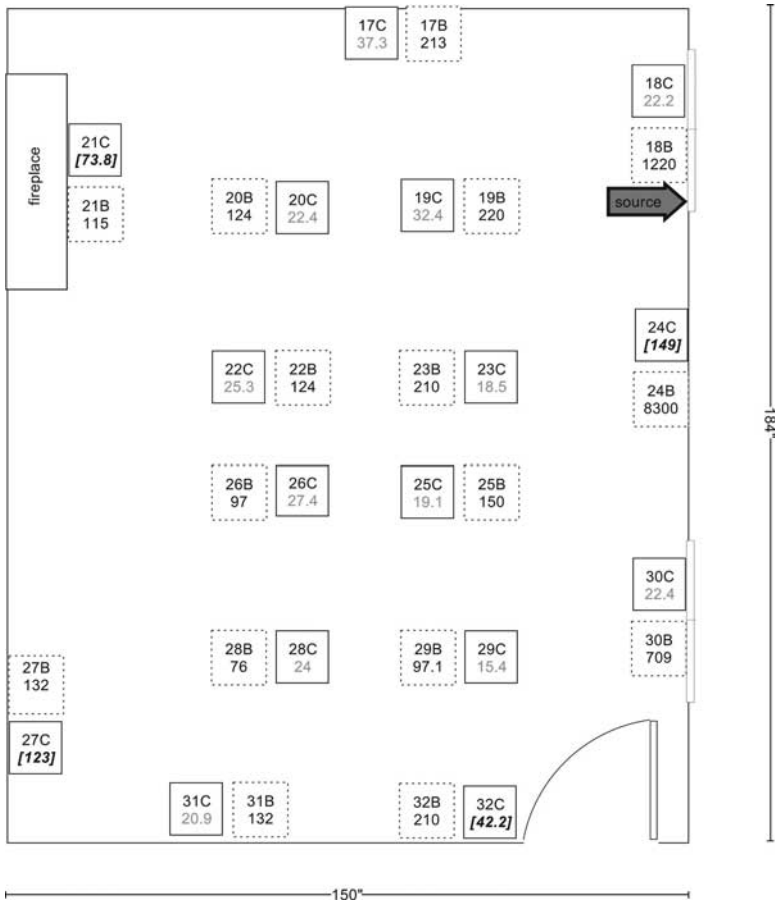

FIG. 1—*Unit 6, Room 2, Living Room, —Leaded Dust Before, —Leaded Dust After.*

placed between them and the adjacent active rooms so that lead dust could migrate from the active into the passive rooms.

Overall, as shown in Table 2, 58% of WALL samples fail clearance, while only 36% of INTERIOR samples fail. There are two possible explanations for the higher post-cleaning loadings on the WALL samples. The first relates to the method of lead dust generation used, i.e., grinding of leaded paint with a mechanical grinder. This tends to produce a plume of dust that spreads across the room from the source. When the plume encounters a vertical surface such as wall, door, cabinet, etc., it descends to the floor at that point and may possibly produce a high lead loading close to the vertical surface. The second possible explanation is that the cleaning process (vacuum/wet mop/vacuum) may be more efficient in the interior of the room than against a vertical surface such as a wall, where access is constrained and dust may accumulate against the vertical surface during the cleaning itself.

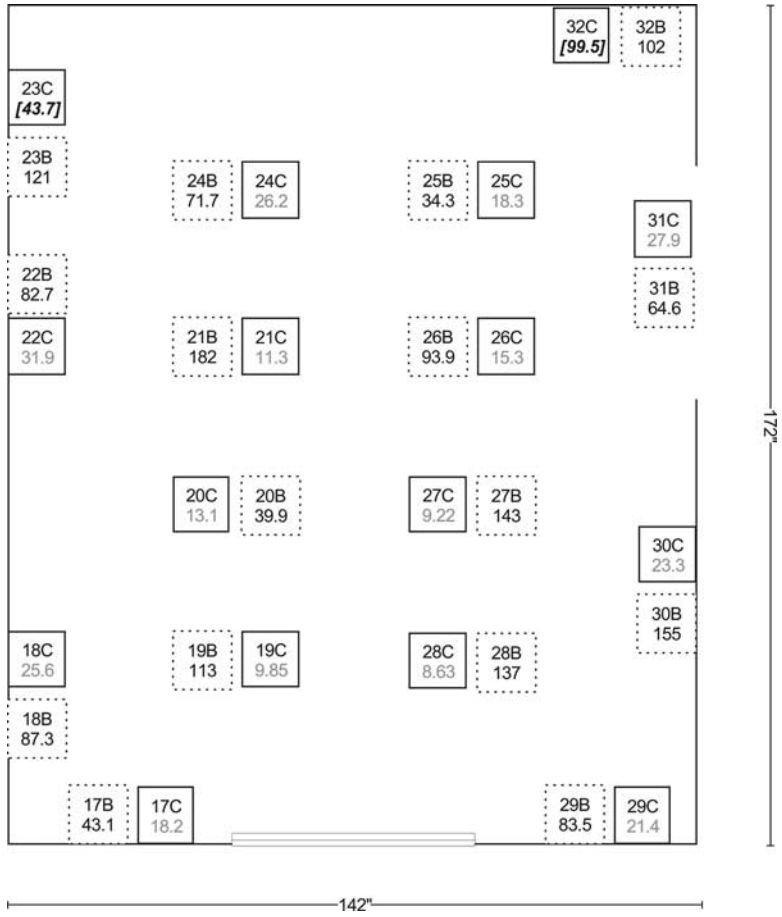

FIG. 2—*Unit 8, Room 2, Front Room, —Leaded Dust Before, —Leaded Dust After.*

The first of these explanations should result in higher WALL lead loadings *pre-cleaning* than INTERIOR loadings, at least in ACTIVE rooms. This is indeed the case. The geometric mean lead loading for INTERIOR samples in ACTIVE rooms pre-cleaning was 293 $\mu g/ft^2$, versus 463 $\mu g/ft^2$ for WALL samples in ACTIVE rooms. The difference was statistically significant ($p = 0.0005$). In PASSIVE rooms, the geometric means were 141 $\mu g/ft^2$ for INTERIOR samples and 122 $\mu g/ft^2$ for WALL samples. This difference was not statistically significant.

To test the second explanation (lower cleaning efficiency near vertical surfaces), it is necessary to control for the higher pre-cleaning loadings near vertical surfaces. Since pre-cleaning and post-cleaning samples were taken in side-by-side pairs, regressions equations were fit for post-cleaning loading as a function of pre-cleaning loading. This gives a measure of the efficiency of cleaning, controlling for the pre-cleaning loading. The fitted equations are as follows (ACTIVE rooms)

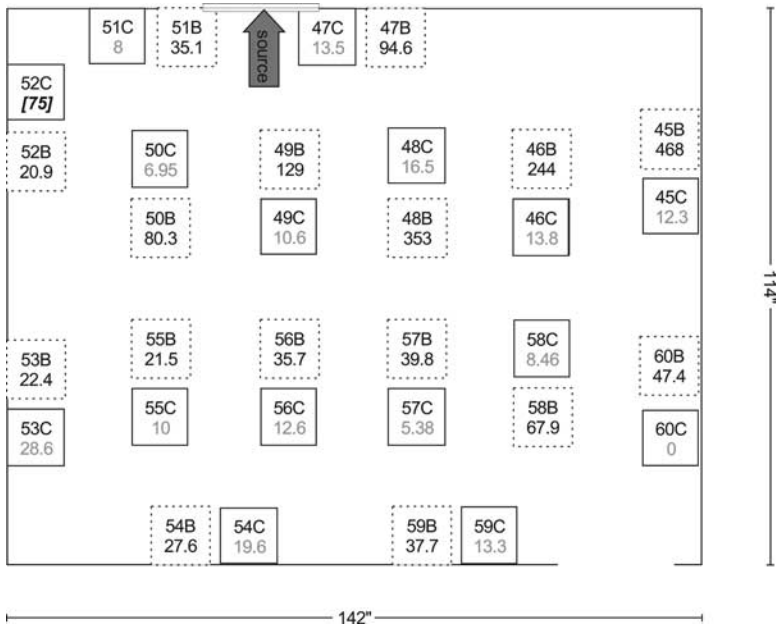

FIG. 3—*Unit 9, Room 4, Bedroom, —Leaded Dust Before, —Leaded Dust After.*

$$\text{INTERIOR SAMPLES} - \text{ACTIVE ROOMS}(N = 210):$$

$$\text{Post}-\text{Cleaning Loading } (\mu g/ft^2) = 2\%(\text{Pre}-\text{Cleaning Loading}) + 43$$

$$\text{WALL SAMPLES}-\text{ACTIVE ROOMS } (N = 183):$$

$$\text{Post} - \text{Cleaning Loading } (\mu g/ft^2) = 6\%(\text{Pre}-\text{Cleaning Loading}) + 92$$

Thus, the cleaning process is greater than 3 times more efficient in the interior of the room as compared to against the wall or other vertical surface. Note that, from these equations, it appears to be difficult to clean to a clearance standard of 40 $\mu g/ft^2$ on average. However, this is an artifact of the study design, since the leaded dust generation process was calibrated to result in levels close to the clearance standard. The dust generation process in the study was considerably more aggressive than one would expect to encounter in a normal renovation, repair or abatement project.

In PASSIVE rooms, the cleaning effect was not observed, with the following fitted equations

$$\text{INTERIOR SAMPLES} - \text{PASSIVE ROOMS}(N = 50):$$

$$\text{Post} - \text{Cleaning Loading } (\mu g/ft^2) = 6\%(\text{Pre}-\text{Cleaning Loading}) + 26$$

TABLE 2—*Number of Samples and Clearance Failures by Room, Unit and Placement (Interior/Wall).*

		Interior		Wall		
Unit	Room	No. Samples	No. Fail	No. Samples	No. Fail	Room Type
2	1	8	1	8	8	Active
2	2	8	4	8	8	Active
2	3	3	0	4	1	Passive
2	4	2	2	3	0	Passive
2	5	8	0	8	4	Active
2	**ALL**	**29**	**7**	**31**	**21**	
3	1	8	8	7	7	Active
3	2	8	8	8	8	Active
3	3	4	1	0	0	Passive
3	4	9	9	11	11	Active
3	**ALL**	**29**	**26**	**26**	**26**	
4	1	8	6	2	2	Passive
4	2	9	2	7	3	Active
4	3	8	1	8	2	Active
4	4	10	0	8	1	Active
4	**ALL**	**35**	**9**	**25**	**8**	
5	1	15	10	9	4	Active
5	2	13	9	7	7	Active
5	3	9	1	7	2	Active
5	**ALL**	**37**	**20**	**23**	**13**	
6	1	10	4	6	5	Active
6	2	10	0	6	4	Active
6	3	6	0	6	2	Passive
6	4	8	1	8	3	Active
6	**ALL**	**34**	**5**	**26**	**14**	
7	1	7	4	7	7	Active
7	2	6	4	2	2	Passive
7	3	8	4	8	8	Active
7	4	5	5	2	2	Passive
7	5	9	9	8	8	Active
7	**ALL**	**35**	**26**	**27**	**27**	
8	1	9	0	7	2	Active
8	2	8	0	8	2	Passive
8	3	8	0	7	3	Active
8	4	5	0	7	0	Active
8	**ALL**	**30**	**0**	**29**	**7**	
9	1	6	0	7	3	Active
9	2	8	0	6	2	Passive
9	3	9	0	8	4	Active
9	4	8	0	8	1	Active
9	**ALL**	**31**	**0**	**29**	**10**	
ALL	**ALL**	**260**	**93 (36%)**	**216**	**126 (58%)**	

$$\text{WALL SAMPLES}-\text{PASSIVE ROOMS}(N=33):$$
$$\text{Post}-\text{Cleaning Loading } (\mu g/ft^2) = 5\%(\text{Pre}-\text{Cleaning Loading}) + 34$$

The difference between ACTIVE and PASSIVE fits was not statistically significant for the PASSIVE rooms. This may indicate that the cleaning effect is only significant when high dust-lead loadings are generated by the project work.

Detection Capabilities of the EPA Clearance Protocol

The EPA clearance protocol (40 CFR Part 745.227) [2] provides for the collection of one floor sample from each of 4 rooms inside the containment area, but does not provide guidance on where the samples should be taken. Clearly, as for any sampling method, the EPA protocol will have a higher chance of detecting clearance failures the higher the percentage of the floor area that is actually above the clearance standard.

Table 3 shows the probability that the EPA protocol will result in clearance failure for each of the units in the study when clearance failure is defined by the EPA protocol as finding *any* of the 4 samples with a lead loading of 40 $\mu g/ft^2$ or higher. Version 1 assumes a sample at a random location in the INTERIOR of each room (or 4 rooms randomly selected if the unit has 5 rooms); Version 2 assumes a random WALL sample instead of an INTERIOR sample. Version 3 assumes that the WALL or INTERIOR selection is made randomly in each room. The table also shows the estimated percentage of the floor area of each unit that is actually at or above the 40 $\mu g/ft^2$ clearance standard. In calculating this percentage, we assumed that 70% of the floor area of each room is INTERIOR (more than 1 ft from any vertical surface) while 30% is WALL (within 1 ft of a vertical surface). This 70-30 split is the median for the rooms in the study.

In each sampled room, and for each sample type (INTERIOR versus WALL), the probability of clearance failure is generally estimated as the proportion of failing samples shown Table 2 for that room and sample type. However, if the proportion of failing samples is either 0 or 1 (no samples fail or all fail), the probability is modified because a true failure probability of exactly 0 or

TABLE 3—*Probability of Detecting Clearance Failure by EPA Protocol (3 Versions).*

Unit	% Area $\geq$ 40 $\mu g/ft^2$	EPA Version 1	EPA Version 3	EPA Version 2
8	7%	0.32	0.46	0.72
9	10%	0.31	0.48	0.83
6	26%	0.56	0.84	0.98
4	28%	0.84	0.87	0.89
2	37%	0.83	0.92	0.99
5	55%	0.91	0.93	0.96
7	82%	>0.995	>0.995	>0.995
3	93%	>0.995	>0.995	>0.995

100% is not likely. When 0 failures are observed in N samples, the failure probability is estimated as $1-(0.5)^{(1/N)}$. For example, if there are 0 failures in 8 samples, the failure probability is estimated as $1-(0.5)^{(1/8)}=0.083$. This value is the true failure probability for which observing 0 failures in N samples has a probability of 50%. When N failures are observed in N samples (100%), the failure probability is likewise estimated as $(0.5)^{(1/N)}$. For example, if 6 failures are observed in 6 samples, the failure probability is estimated as $(0.5)^{(1/6)}=0.891$.

In Units 3 and 7, where over 80% of the floor area has a lead loading of 40 $\mu g/ft^2$ or greater, all 3 versions of the EPA Protocol have essentially 100% detection capability. In the other units, where the floor area with a lead loading of 40 $\mu g/ft^2$ or greater ranges from 7% to 55%, Version 2 of the EPA Protocol (WALL samples only) has the best detection capability, followed by Version 3 (random selection of WALL versus INTERIOR in each room) and then Version 1 (INTERIOR samples). In the units with lowest percentage of floor area with a lead loading of 40 $\mu g/ft^2$ or greater (Units 6, 8, and 9), the difference in capability of detecting clearance failure between the 3 versions of the EPA Protocol is substantial. For example, in Unit 6, the WALL sampling version has 98% probability of detecting clearance failure, while the INTERIOR version has only a 56% chance of detection (not much better than 50-50). Thus, it is clear than *the exact method by which the EPA protocol is implemented can have a significant impact on the likelihood of detecting clearance failures, particularly when the percentage of floor area in the unit with a lead loading of 40 μg/ft* [2] *or greater is less than 50%.*

Modifications to the EPA Clearance Protocol

Two potential modifications to the EPA Clearance Protocol are (a) increase the number of samples or (b) take composite samples.

(a) Increased Sampling—One possible modification to the EPA Protocol would be to take 2 samples per room, rather than 1. This would increase the analysis cost for clearance sampling, but not the far larger part of the cost, which is the labor of the Risk Assessor for travel, on-site time, and reporting. Thus, the overall impact on the cost of clearance would be modest. Table 4 shows the detection probabilities for the same 3 versions of the EPA Protocol previously discussed for Table 3.

The two-sample-per-room EPA Protocol increases the detection capability for all units and versions, of course. For Version 2 (WALL samples) the detection capability is over 90% for all units, even when the % of floor area with a lead loading of 40 $\mu g/ft^2$ or greater is low. However, detection capability is still weak for Units 8 and 9 using Version 1 (two INTERIOR samples randomly selected) and Version 3 (where a WALL or INTERIOR is randomly selected twice to get the 2 samples).

(b) Composite Sampling—An alternative approach to improving the detection capability of the EPA Protocol is to take composite samples. To evaluate the potential use of collecting composite samples, results from the individual

TABLE 4—*Probability of Detecting Clearance Failure by EPA Protocol (3 Versions) With Two Samples per Room.*

Unit	% Area ≥ 40 $\mu g/ft^2$	EPA Version 1	EPA Version 3	EPA Version 2
8	7%	0.54	0.71	0.92
9	10%	0.52	0.73	0.97
6	26%	0.81	0.97	>0.995
4	28%	0.97	0.98	>0.995
2	37%	0.97	0.99	>0.995
5	55%	0.99	>0.995	>0.995
7	82%	>0.995	>0.995	>0.995
3	93%	>0.995	>0.995	>0.995

wipe samples collected in this evaluation were mathematically summed. Consider taking 4 wipe samples in each room to form a composite of the 4 wipe samples which is extracted and analyzed by the laboratory as a single sample. While laboratory analysis of 4-wipe composites may be slightly more costly than single-wipe samples, the overall cost impact of compositing is minimal. The lead loading for a composite sample is the arithmetic average lead loading for the 4 samples making up the composite. Therefore, the effect of compositing strategies can be evaluated mathematically using the individual lead loadings found in each room of the study. A logical method of compositing would be to composite 2 randomly-selected INTERIOR with 2 randomly-selected WALL samples in each room.

The EPA regulations at 40 CFR 745 [2] have established a clearance standard for floors utilizing composite wipe sampling as 40 $\mu g/ft^2$ divided by half the number of sub-samples in the composite sample, i.e., 20 $\mu g/ft^2$ for a 4-wipe composite. In other words, a 4-sample composite must meet a standard numerically twice as stringent as a single sample. The probability that a 4-sample composite taken in a room of the study (2 INTERIOR and 2 WALL samples chosen randomly from the study samples) will exceed the 20 $\mu g/ft^2$ standard can be evaluated mathematically for each room. The probability of a clearance failure in a given room, P(F), is given by the ratio

P(F) = [Number of 4-sample composites $\geq$20-$\mu g/ft^2$/[Total number of 4-sample composites].

Consider, for example, Unit 9, Room 4 (shown in Fig. 3). The lead loadings for the individual post-cleaning samples are shown in color (green for samples <40 $\mu g/ft^2$ and red for samples ≥ 40 $\mu g/ft^2$). The individual values are such that, if sample 52C is chosen as one of the 4 samples to be composited, then the composite value will always exceed 20. Conversely, if sample 52C is *not* chosen, then the largest possible composite sample value, using 2 WALL and 2 INTERIOR samples, is $(28.6 + 19.6 + 16.5 + 13.8)/4 = 19.6 < 20$ $\mu g/ft^2$. This value is obtained when samples 53C, 54C, 48C, and 46C are chosen. Thus, in the case of Unit 9, Room 4, the probability that a composite sample $\geq$20-$\mu g/ft^2$ is obtained is exactly the probability that 52C is chosen as one of the two WALL samples in the 4-composite sample. Since there are 8 WALL samples total, this probability

is $1-(7^{*}6)/(8^{*}7)=0.25$. A similar calculation process was performed for all the rooms in the study, and the resulting probabilities combined to obtain, for each unit, the probability that the 4-composite sample in at least one room of the 4 sampled would have a value of 20 $\mu g/ft^2$ or greater. This probability was at least 99% in every unit. Thus, *using a composite of 4 samples in each room in the unit (2 WALL and 2 INTERIOR selected randomly), the probability of detecting clearance failure was 99% or greater in every unit*.

It is possible for a 4-wipe composite sample to result in a "false positive," i.e., a composite value ≥ 20 $\mu g/ft^2$ when all 4 samples comprising the composite are actually below the 40 $\mu g/ft^2$ standard for individual samples. In this study, Unit 8, Room 4, was the only room where *all* individual floor samples were below 40 $\mu g/ft^2$. In this case, the maximum possible value for a 4-sample composite was $(19.5+16.6+16.5+14.1)/4=66.7/4=16.7<20$ $\mu g/ft^2$. Thus, no composite samples fail, and there are no false positives. In all other study rooms, whenever a composite fails (even if no individual sample in that particular composite is ≥ 40 $\mu g/ft^2$) there is in fact some floor area ≥ 40 $\mu g/ft^2$ in the room. The study therefore provides some limited evidence that, in practice, the distribution of post-cleaning floor dust lead levels may tend to limit false positives for the 4-sample composite protocol.

Conclusions

The main conclusions are:

(1) Floor dust-lead along the perimeters of rooms was three times more difficult to clean than dust-lead from the interiors.

(2) The exact method by which the EPA protocol is implemented can have a significant impact on the likelihood of detecting clearance failures, particularly when the percentage of floor area in the unit with a lead loading of 40 $\mu g/ft^2$ or greater is less than 50%.

(3) Clearance failure is much more likely for floor samples taken near the walls or other vertical surfaces in a room, so that clearance sampling protocols should emphasize perimeter sampling over interior sampling.

(4) Composite sampling provides a very reliable method of detecting clearance failure without significantly increasing the cost of clearance sampling. Using a composite of 4 samples in each room in the unit (2 WALL and 2 INTERIOR selected randomly), the probability of detecting clearance failure was 99% or greater in every unit. The conduct of clearance as defined by EPA does not exclude the use of compositing wipe samples for lead determinations. However, at the time that this article was written, analysis of composite dust wipes for lead is not a part of the NLLAP. Therefore, there may be some difficulty in obtaining lead analysis for composite wipe samples under this accreditation program.

Acknowledgments

Financial support from the U.S. Department of Housing and Urban Development Office of Healthy Homes and Lead Hazard Control under Contract

Number C-OPC-21358 is gratefully acknowledged. The writers also express appreciation to John Zilka for providing field supervision and to Duncan Holmes and Terry Phillips for their efforts in collecting field data. Particular thanks are extended to those responsible for providing access to suitable units meeting the needs of this study. For Youngstown units, we are indebted to Gary Singer, Diana Seydlorsky, and Connie Thompson of the Mahoning County Healthy Homes/Lead Hazard Control Program, and to Gary Crim, landlord. For Erie units, we thank Jane Grogan, Bill Hollenbeck, Sherri Schultz, and Tim Heirls of the Redevelopment Authority of the City of Erie (R.A.C.E.). Finally, thanks are extended for the risk assessment support services provided by Daniel J. Casciato Jr. and Patrick Gool of the Real Estate Inspection Service.

References

[1] Title 24, *Code of Federal Regulations*, Part 35-Lead-Based Paint Poisoning Prevention in Certain Residential Structures.

[2] Title 40, *Code of Federal Regulations*, Part 745-Lead-Based Paint Poisoning Prevention in Certain Residential Structures.

[3] "Standard Specification for Wipe Sampling Materials for Lead in Surface Dust," *ASTM E1792-03*.

[4] U.S. Dept. of Housing and Urban Development, "Guidelines for the Evaluation and Control of Lead-Based Paint Hazards in Housing," Washington, DC, June 1995 as amended.

[5] "Standard Practice for Collection of Settled Dust Samples Using Wipe Sampling Methods for Subsequent Lead Determination," *ASTM E1728-03*.

[6] "Standard Practice for Ultrasonic Extraction of Paint, Dust, Soil, and Air Samples for Subsequent Determination of Lead," *ASTM E1979-04*.

[7] "Standard Test Method for Determination of Lead by Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-AES), Flame Atomic Absorption Spectrometry (FAAS), or Graphite Furnace Atomic Absorption Spectrometry (GFAAS) Techniques," *ASTM E1613-04*.

[8] U.S. Environmental Protection Agency, "EPA National Lead Laboratory Accreditation Program Laboratory Quality System Requirements," *Rev. 3.0*, Washington, DC (July 5, 2007).

[9] *American Industrial Hygiene Association (AIHA) Proficiency Analytical Testing Programs (PATP), LLC*, 2700 Prosperity Ave., Fairfax, VA.

Reprinted from JAI, Vol. 8, No. 3
doi:10.1520/JAI103465
Available online at www.astm.org/JAI

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Pilot Evaluation for Lead-Based Paint Proficiency Testing of Field Portable XRF Instruments

ABSTRACT: A proficiency testing procedure for evaluating the accuracy of lead measurements taken by certified lead-based paint inspectors using portable X-ray fluorescence (XRF) instruments was developed and pilot tested. XRF instruments are widely used for inspection of housing units to determine the presence or absence of lead-based paint. Previous research showed variability between different XRF instruments when used to conduct lead-based paint inspections. Due to concerns over potential errors in inspection reports resulting from biased XRF measurements, a proficiency testing procedure was pilot tested to provide entities, such a state or federal regulators, a means of evaluating the instrumentation being used for lead-based paint inspections.

KEYWORDS: XRF, proficiency, lead-based paint

Introduction

Detection of lead-based paint (LBP) in housing, defined in regulations as a LBP inspection, is often performed using a field-portable X-ray fluorescence (XRF) instrument. These instruments are factory calibrated, and are often used during LBP inspections with few or no quality control procedures to verify the ability

Manuscript received October 8, 2010; accepted for publication February 17, 2011; published online March 2011.
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Cite as: White, K. T., Dewalt, F. Gary, Cox, D. C., Schmehl, R., Friedman, W. and Pinzer, E. A., "Pilot Evaluation for Lead-Based Paint Proficiency Testing of Field Portable XRF Instruments," *J. ASTM Intl.*, Vol. 8, No. 3. doi:10.1520/JAI103465.

of the XRF to provide accurate results. Data from previous research 1, referred to here as the baseline study, show that field-portable XRF instruments have considerable variability and sometime show bias when used to measure leaded-paint test films placed over commonly evaluated building material substrates. This implies that some XRF instruments may not make determinations with sufficient accuracy to correctly identify the presence or absence of LBP in coatings, and that some XRF instruments perform differently than others of the same make/model. LBP inspections are applied to a variety of needs that include investigations seeking to identify the causes of lead poisoning of children (often referred to as elevated blood lead (EBL), investigations). In most states, LBP inspections are regulated by state lead hazard control program offices (however titled). Given the importance of LBP inspections, some governmental agencies regulating their conduct have expressed an interest in a practical method of evaluating the performance of XRF instruments being used by lead inspectors. This process, called "proficiency testing," is a common feature of laboratory accreditation programs [2] such as the National Lead Laboratory Accreditation Program (NLLAP) [3]. However, proficiency testing is difficult to apply to in-field testing operations. A procedure for evaluating the performance of XRF instruments was developed for the U.S. Department of Housing and Urban Development (HUD) Office of Healthy Homes and Lead Hazard Control (OHHLHC) and pilot tested by a state lead program office to determine the practically for widespread use. A portable set of test samples, developed to mimic real-world building substrates having lead-containing coatings, was obtained by the state on loan from the OHHLHC. These materials were provided to a state lead hazard control program office along with specific written instructions (a program guidance document) for the conduct of proficiency testing and collection and processing of data.

Materials and Methods

Test Samples

The test samples, referred to as the National Lead Assessment and Abatement Council (NLAAC) (now the Lead and Environmental Hazards Association) test films, were obtained by loan from OHHLHC under a custodian transfer agreement from the permanent custodian, QuanTech, Inc., to an agreed upon custodian at the participating state lead program office. These test samples, owned by HUD, were developed under a grant to NLAAC in 1995 for evaluation of field-portable XRF instruments. The lead concentrations in these paint film samples are closely held to prohibit potential bias and outside influence when conducting evaluations of XRF instruments. The transfer agreement defines the handling and control of the testing materials so that the blind nature of the lead values of the paint films is maintained for future use. Test samples were returned to HUD custody after the completion of testing. The test samples are comprised of three sets of materials: Paint films containing known amounts of lead, substrate test blocks, and test stands.

Paint Films—"Stock solutions" of leaded paint were used to create paint films with different lead concentrations by mixing a white lead paint (containing approximately 53 % lead by weight) with commercially available household semi-gloss alkyd paint. The white lead paint was made from a mixture of boiled linseed oil, raw linseed oil, white lead, and a small amount of mineral spirits. The paint films were prepared by spreading the stock solutions on Mylar sheeting using a drawdown blade. Mylar was selected because of its lack of porosity, uniform thickness, and negligible impact to XRF measurements when used as a support substrate. This drawdown technique was used to provide films having uniform thickness, width, and length dimensions. After drying, uniformity of thickness was verified across the film sheets using a digital thickness gauge (Ultra-Digit Mark IV available from Fred V. Fowler Co., Inc., Newton, MA). Using a paint roller, varying numbers of layers of a non-lead containing, gray-tinted commercially available household semi-gloss alkyd paint was randomly added to the various film sheets containing the different lead levels. Coupons cut from the finished film sheets were labeled on an underside corner with a unique identification number and heat sealed between polyethylene sheeting for protection against potential inadvertent handling-related damage. Lead levels in the test films were determined by laboratory analysis of multiple samples collected from the films sheets immediately adjacent to each coupon. The estimated uncertainty in the lead level of a given test film is about 5 % or less [4]. All the NLAAC test films look identical in both color and shape when presented to a participant for testing. A total of 16 different test films are included in a set. Six total sets have been produced. Only one set was used in this work.

Substrate Test Blocks —Test blocks are made from construction materials commonly encountered in housing upon which paint film(s) may be overlaid to simulate painted surfaces. The test blocks are labeled "wood," "metal," "concrete," "plaster," "wallboard," "brick," and "cinder block," as appropriate.

Test Stands —These are used to hold the test films in place on top of the substrate test blocks. The test stands are constructed from 5 U.S. gal plastic food container buckets. The buckets are filled with expanded polystyrene foam blocks to eliminate potential background interference. The distance from the bottom of a substrate test block to the bottom of the bucket is at least 10 in.

Data Collection and Reporting

Included with the loan of the test samples is proficiency testing program guidance document. This guidance document contains instructions on recruiting, test scheduling, handling of test samples materials, testing by XRF operators, data collection, and submission of data to the permanent test sample custodian for analysis and reporting. The "third party" data analysis step performed by QuanTech is required because the identity of the test samples is blind to the participating state lead program.

Data collection consisted of having the state lead program office schedule XRF operators (state-licensed LBP Inspectors and Risk Assessors) to come to a

specified location and conduct lead-content measurements on a total of 17 test samples consisting of test films (16 NLAAC films plus NIST SRM 2573 at 1.0 mg/cm^2) (National Institute of Standards and Technology) prearranged on substrate test blocks placed in the test stands. The pairing of paint films and substrate blocks for each test sample is defined in the program guidance document. The pairings match those used in the baseline study [1] to allow for statistical comparisons to this study. The XRF operators conducted testing for lead in paint according to their company protocols and reported lead content in units of milligrams of lead per square centimeter (mg/cm^2) for each test sample. The participating state lead program representative consolidated and then transferred the collected data to QuanTech for analysis and reporting. Individual performance results were reported to each XRF instrument operator and collectively to the participating state. All of these reports were formatted and delivered in a manner to protect the operator performance, paint film identity, and paint film lead content.

Data Analysis Using the Baseline Study

Data from the baseline study [1] was used as a benchmark for gauging the performance of the individual XRF instruments evaluated by the participating state lead program. As shown in Table 1, four principal types of XRFs were represented among the 94 inspectors involved in the baseline study: RMD, NITON, Scitech, and PGT. Other instruments were represented by at most one or two inspectors each.

Some of the inspectors shared instruments such that of the 94 inspectors, only 71 distinct instruments were used as indicated in the last column of Table 1. A substantial database of measurements on each of 16 NLAAC films was generated in the baseline study for the four instruments most commonly used. Thus, for these four types of instruments, it is possible to determine whether an

TABLE 1—*XRF instruments tested during the baseline study.*

Manufacturer/ Model	Total Number of Separate Tests on NLAAC Test Films	Total Number of Unique XRF Instruments Making Separate Tests on NLAAC Test Films
RMD[a]	45	31
NITON[b]	23	19
Scitec/MAP4[c]	10	12
PGT/XK-3[d]	9	6
Other	7	3

[a] RMD (Radiation Monitoring Devices, Inc., 44 Hunt Street, Watertown, MA 02172).
[b] NITON (NITON, LLC, 900 Middlesex Turnpike, Building #8, Billerica, MA 0182).
[c] Scitec model MAP4 (EDAX Portable Products Division, 415 North Quay, Kennewick, WA 99336).
[d] PGT model XK-3 (Princeton Gamma-Tech, Inc., 1026 Route 518, Rocky Hill, NJ 08553).

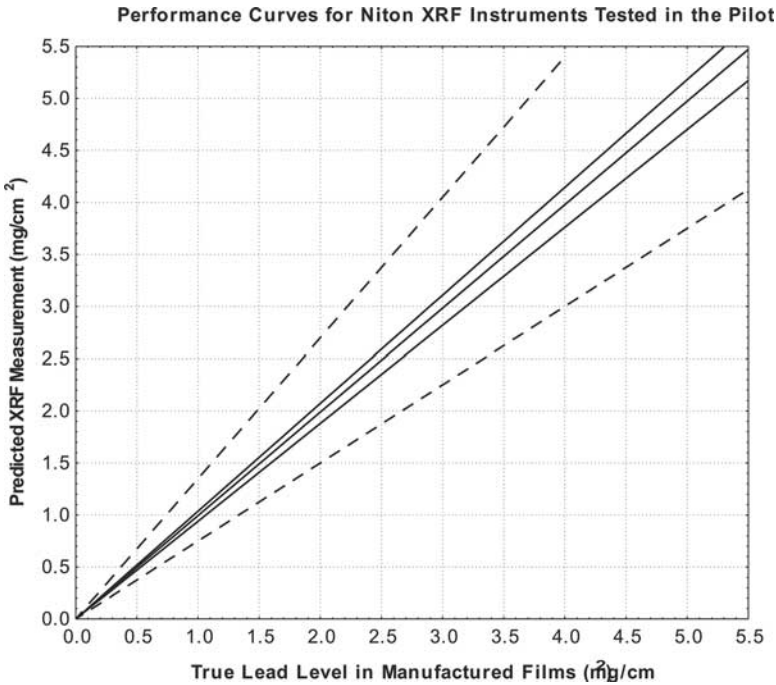

FIG. 1—*Linear regression fits for NITON pilot instruments as compared to NITON instruments used in the baseline study. Dashed lines represent two standard deviations from the mean for the NITON instruments evaluated in the baseline study. Solid lines represent the performance of the three NITON instruments evaluated in this study.*

individual instrument of the same type generates measurements consistent with those of that type seen in the baseline study. A regression analysis methodology was used to carry out this determination. In this pilot test, only NITON and RMD XRF instruments were tested. Therefore, data analysis was limited to these two instruments.

The baseline study database contains measurements from 19 distinct (different serial numbers) NITON and 31 distinct RMD instruments. In a number of cases, the same instrument was used by more than one operator with up to a maximum of five different operators using one particular RMD instrument. For analysis purposes, all data from the same instrument were combined, resulting in different sample sizes for the various instruments. Data on NLAAC films with true lead levels in excess of 5 mg/cm^2 were excluded from analyses of the NITON instruments because such instruments do not read above 5 mg/cm^2.

Test results on the two lowest lead level NLAAC films (having less than 0.07 mg/cm^2 of lead) for all of the NITON and RMD instruments in the baseline study were not statistically different from zero. Therefore, a zero-intercept regression line fitting of the data points was used to relate response plots (XRF instrument response to the accepted (true) lead levels in the tested NLAAC test films). Analysis as to the acceptability of the pilot test instrument to achieve the

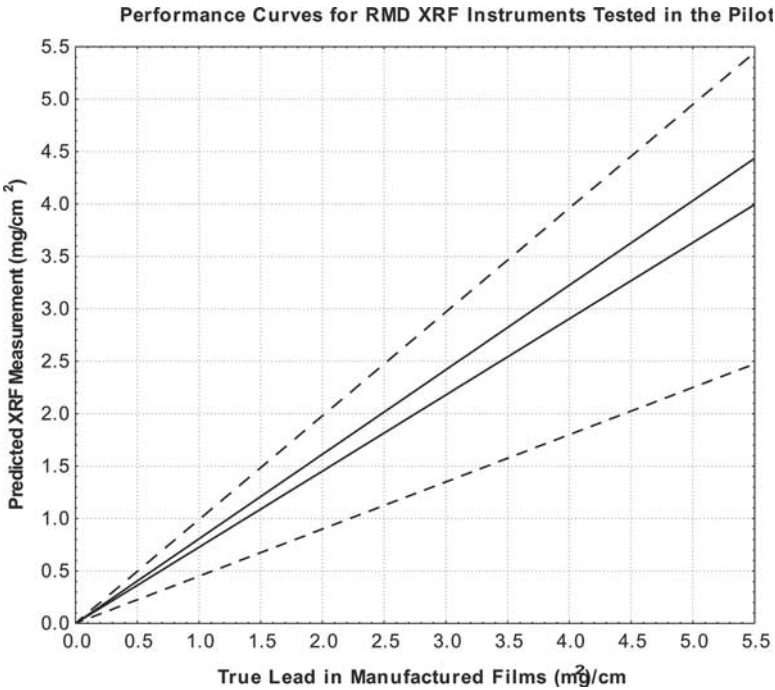

FIG. 2—*Linear regression fits for RMD pilot instruments as compared to RMD instruments used in the baseline study. Dashed lines represent two standard deviations from the mean for the RMD instruments evaluated in the baseline study. Solid lines represent the performance of the two RMD instruments evaluated in this study.*

performance shown in the baseline study was performed visually from plots of the tested XRF against plots of that XRF type using data from the baseline study.

Linear regression fits of data from the baseline tested NITON instruments had R^2 values greater than 0.9. For the RMD instruments, R^2 were greater than 0.8. The slope of the zero-intercept linear regression for the NITON in the baseline study data ranged from 0.85 to 1.49 with a mean of 1.05 and a standard deviation of 0.15. For the RMD, the range was 0.49 to 1.15 with a mean of 0.72 and a standard deviation of 0.14. For purposes of comparison to the pilot tested instruments, a range of regression slopes was plotted equal to the mean plus and minus two standard deviations. This creates two line plots bracketing approximately 95 % of the baseline study population. This range of slopes for use as the benchmark is 0.75 to 1.35 for the NITON, and 0.45–0.99 for the RMD.

Results

In this pilot, data were collected on five XRF instruments: Three manufactured by NITON, and two by RMD. Fourteen measurements were available for each of the three NITON instruments in the pilot. One RMD had 16 measurements and the other, which was run twice through the sample set, had 32 measurements.

Figure 1 shows the estimated regression lines for the three NITON instruments compared to the range for the baseline study instruments. Figure 2 shows the equivalent data for the two RMD instruments.

Conclusions

Figures 1 and 2 show that all XRF instruments tested in this pilot performed within two standard deviations from the mean for like brands evaluated in the baseline study. Therefore, the XRF instruments in the pilot have performed no differently than those previously evaluated [1]. This implies that these instruments should be capable of detecting the presence or absence of LBP as well as those evaluated previously. Further, the proficiency testing program pilot tested here was found to be suitable for use by organizations for assessing the performance of XRF instrumentation used for LBP detection. The data presented here can serve as a tool for setting acceptance limits for XRF instrument performance.

Acknowledgments

Financial support from the U.S. Department of Housing and Urban Development Office of Healthy Homes and Lead Hazard Control under Contract Nos. C-OPC-21358 and C-PHI-01043 is gratefully acknowledged. The authors also express appreciation to Mr. William Dundulis from the State of Rhode Island and Providence Plantations for his assistance during conduct of this work. Mention of company or product names does not constitute endorsement. This article was prepared by U.S. Government employees and contractors as part of their official duties and legally may not be copyrighted in the United States of America.

References

[1] Dewalt, G., Cox, D., Friedman, W., and Jacobs, D. E., "Field Evaluation of Lead-Based Paint Inspections Final Technical Report," Vols. 1–2, U.S. Department of Housing and Development Office of Lead Hazard Control, Washington, D.C., September 3, 1998, available from the Federal Lead Clearinghouse, 1-800-424-LEAD.

[2] ISO/IEC 17025, 2005, "General Requirements for the Competence of Testing and Calibration Laboratories," International Organization for Standardization (ISO), Geneva, Switzerland.

[3] U.S. Environmental Protection Agency, "EPA National Lead Laboratory Accreditation Program Laboratory Quality System Requirements," Rev. 3.0, U.S. Environmental Protection Agency, Washington, D.C. July 5, 2007.

[4] QuanTech, Inc., "Confidential (Proprietary) Report on the Preparation of Proficiency Testing Samples for the NLAAC XRF Operators Registry," U.S. Department of Housing and Development Office of Lead Hazard Control, Washington, D.C., April 1996.

BERYLLIUM

Reprinted from JAI, Vol. 8, No. 8
doi:10.1520/JAI103411
Available online at www.astm.org/JAI

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Beryllium Measurement by Optical Fluorescence in Samples Contaminated by Strongly Fluorescent Impurities

ABSTRACT: A fluorescence method for determination of beryllium and beryllium oxide particles has been approved as a standard test method by ASTM International and the National Institute for Occupational Safety and Health. The procedure involves dissolution of samples in ammonium bifluoride solution and adding a small aliquot of extract to a basic hydroxybenzoquinoline sulfonate fluorescent dye and measuring its fluorescence. This method is specific to beryllium and is not affected by the presence of other metals. However, the results from such a method may be compromised by organic fluorescent impurities which have optical characteristics similar to the beryllium-bonded hydroxybenzoquinoline sulfonate. This study demonstrates that such impurities can be effectively removed by activated charcoal without compromising the sensitivity of the method or any other test attributes.

Introduction

Analysis of beryllium by fluorescence is a well established method [1–4] and has been promulgated as a standard test procedure for determining beryllium in particles collected on wipe samples, air filters and soil samples. These methods are available as American Society of Testing Materials International (ASTM) methods D7202 [5] and D7458 [6], and National Institute for Occupational

Manuscript received October 4, 2010; accepted for publication June 9, 2011; published online July 2011.
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Cite as: Agrawal, A., Adams, L., Agrawal, A., Cronin, J. P., Lopez Tonazzi, J. C. and Duran, B., "Beryllium Measurement by Optical Fluorescence in Samples Contaminated by Strongly Fluorescent Impurities," *J. ASTM Intl.*, Vol. 8, No. 8. doi:10.1520/JAI103411.

Safety and Health (NIOSH) 7404 and 9102 [7]. Alternative methods used to determine beryllium are graphite furnace atomic absorption spectrometry (GFAAS), inductively coupled plasma atomic emission spectrometry (ICP-AES) and inductively couple plasma mass spectrometry (ICP-MS) [8,9]. Compared to the other methods, the fluorescence method is not sensitive to interference by other metals present [10] and has beryllium detection limits comparable to the most sensitive method using ICP-MS. However, no work has been reported on the interference effects of organic fluorescence impurities in samples where beryllium determination is carried out by fluorescence.

When the impurities have an emission and excitation spectrum overlapping with the HBQS-Be spectrum, these will manifest as a positive bias in results as increased beryllium concentration. In this study we investigated a method to remove the fluorescent impurity by deliberately contaminating the sample with succinimidyl ester (Marina Blue) [11], which strongly overlaps both with the excitation and the emission spectrum of HBQS in the presence of beryllium. In this work, the initial effort was placed on developing the method, where Marina Blue contamination was successfully removed. The new method was then evaluated on two types of samples. The first evaluation was on soil and sediment standard reference materials (SRMs) that were suspected of having organic fluorescent impurities. The second evaluation was on individually packaged wipes which are used for beryllium surface sampling.

Methods

Materials

Three types of soils and sediments were evaluated, which were SRMs from the National Institute of Standards and Technology (NIST, Gaithersburg, MD). These were SRM 2702 (Waterway sediment), SRM 1944 (Marine sediment) and SRM 2710 (Montana soil). Four types of wipes were compared: Palintest Dust Wipes (available from Palintest USA, Erlanger, KY), Ghost wipes (from Environmental Express, Mount Pleasant, SC), Lead Wipe (Lynx Products, Thorofare, NJ); Whatman 541 and 0.8 μm pore size MCE (mixed cellulose ester) filters (available from Fisher Scientific, Pittsburgh, PA). The first three were prewetted and individually wrapped, while Whatman 541 and MCE filters came in a dry pack in bundles of 100 wipes. Whatman 541 filters are used as wipes for surface sampling and MCE filters are used in cassettes for air sampling. Marina Blue (succinimidyl ester was obtained from Invitrogen (Carlsbad, CA). Two varieties of activated charcoals, Darco G60 and acid washed with hydrochloric acid were obtained from Sigma-Aldrich (Milwaukee, WI). For filtering solutions, the syringe filters used were 0.2 μm hydrophilic polypropylene (GHP) filters (obtained from VWR International).

For determination of beryllium by fluorescence the air or wipe samples (e.g., using 37 mm MCE air filters or Whatman 541 wipes) were first subjected to a dissolution process by placing them in 5 ml of 1% ammonium bifluoride (ABF) and heating to 80 to 90°C for at least 30 mins. Fluorescence measurement solutions were then prepared in cuvettes by adding 0.1 ml of the

dissolution solution to 1.9 ml of a detection solution (20x dilution) containing 1.1mM HBQS, 1mM ethylenediamine tetraacetic acid and 100mM L-Lysine monochloride with the solution pH adjusted to 12.85. In this study, the fluorescence data for beryllium quantification was performed using a Turner BioSystems Modulus fluorometer (Sunnyvale, CA) with a bandpass filter for excitation and emission. The filter transmission characteristics for excitation were 365 ± 10 nm and for emission 480 ± 5 nm, respectively. The fluorometer was calibrated using beryllium standard solutions supplied by Berylliant Inc, Tucson, AZ. The standards had beryllium concentrations of 0, 10, 40, 200 and 800 μg/L. When these standards are mixed with HBQS (20X dilution), the beryllium concentrations were 0, 0.5, 2, 10 and 40 ppb in the measurement solution. These standards resulted in a linear calibration of the fluorometer with a 0.99999 correlation coefficient or better.

For experiments required to investigate spectral characteristics, a Shimadzu RF-5301 PC Spectrofluorophotometer (Columbia, MD) was employed. For soils and sediments the procedure was as described in ASTM D7458, where the dissolution of 0.5g of sample was conducted in 50 ml of 3 wt% ABF solution for 40 hs at 90°C. These extracts were then analyzed using 20X dilution process as described above. In this study we considered several types of wipes which have been traditionally used for beryllium sampling. Since some of these are large in size we had to use 20 ml of 1% ABF dissolution solution. For detection, 0.4 ml of this solution was mixed with 1.6 ml of the HBQS dye solution (5X dilution process [2]). This allowed us to compensate the sensitivity loss that was encountered by using larger amount of the dissolution solution (20 ml versus 5 ml).

Initial experiments were conducted to develop the method, where samples spiked with Marina Blue were evaluated. The protocols for the amount of charcoal required, the sequence of experimental steps and processing conditions were established. The fluorescent dye, HBQS, was prepared in house [12].

Results and Discussion

The unique aspect of the fluorescence method to detect beryllium is the use of the fluorescence dye (HBQS) which specifically binds to beryllium. The phenolic group binds strongly to beryllium where the six member ring has the ideal distance between O–O or N–O for chelating Be [10]. The chemical structure of HBQS is shown in Fig. 1.

Figure 2 shows the excitation spectra for the measurement solution with no beryllium and with specific amounts of beryllium. This spectrum shows relative intensity requirements at each wavelength for a equivalent amount of signal at 475 nm. Figure 3 shows the fluorescence spectra for all of these samples at an excitation wavelength of 365 nm. The maximum fluorescence emission is seen at 475 nm for the dye bound to beryllium. A tightly bound hydrogen bonded proton leads to a weak triplet emission at 580 nm for the dye solution. When the proton is displaced by a metal such as beryllium, fluorescence emission is observed at 475 nm [10].

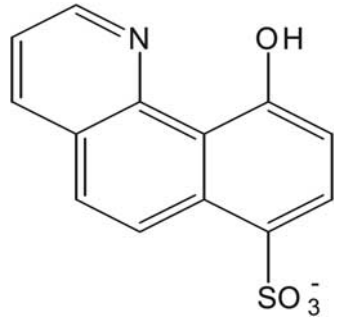

FIG. 1—*Structure of hydroxybenzoquinoline sulfonate (HBQS) fluorophore.*

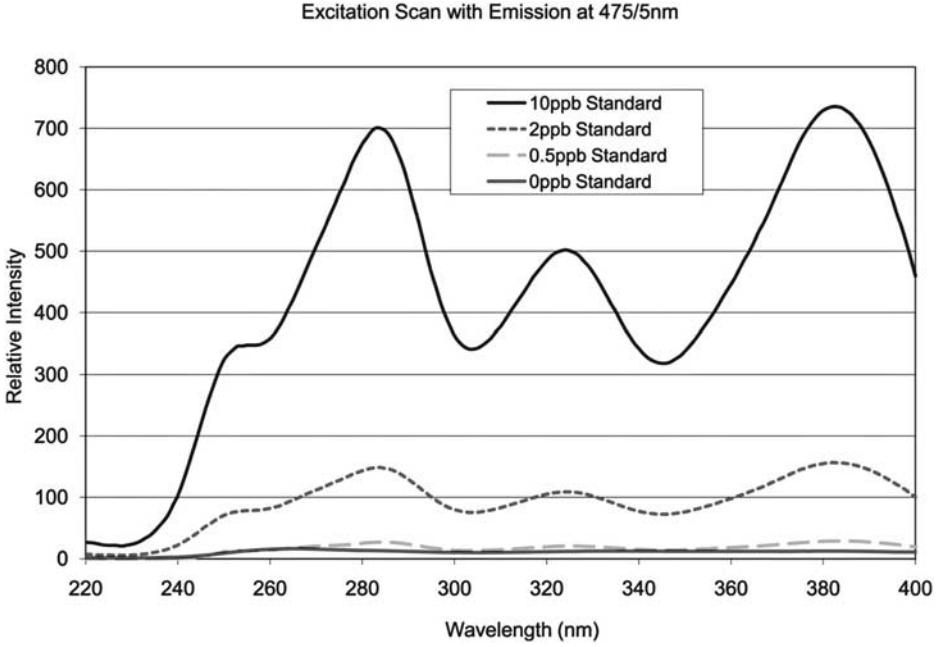

FIG. 2—*Excitation scan, Intensity of radiation required to obtain equivalent fluorescence intensity of hydroxybenzoquinoline sulfonate (HBQS) dye at 475 nm with different concentrations of beryllium acetate standard.*

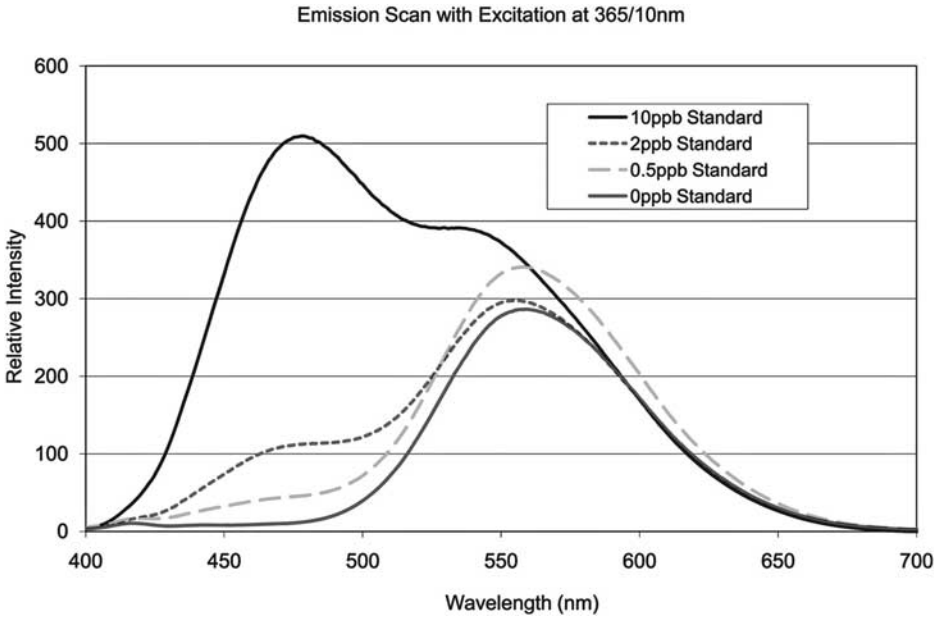

FIG. 3—*Emission spectra (fluorescence intensity) of hydroxybenzoquinoline sulfonate (HBQS) dye with different concentrations of beryllium acetate standard. Excitation wavelength was 365 nm.*

Samples Spiked with a Known Fluorophore

To model the effect of a fluorescence impurity, it was desirable to select a fluorescence dye which had similar emission and excitation characteristics at 365 and 475 nm as the HBQS dye. One such material is Marina Blue (succinimidyl ester). The dye has a molecular weight of 367.26 g/mol and an excitation/emission wavelength of 365/460 nm. Its chemical structure is shown in Fig. 4.

Figure 5 shows the effect of adding Marina Blue to beryllium-containing samples with HBQS dye. The figure shows the emission spectra for a 0.5 ppb beryllium standard with and without Marina Blue and the spectra of Marina Blue in the detection solution without any added beryllium. The addition of Marina Blue causes an increase in the emission signal at 475 nm and interferes positively with the signal due to HBQS chelating with beryllium, resulting in an inflated value for the beryllium concentration. The raw fluorescence intensity value at 475 nm for the 0.5 ppb beryllium standard went from 70.4 to 130.7 with the addition of Marina Blue.

Impurities in solutions can be removed chemically through the use of; for example, oxidants/reductants, bleaches, selective precipitation or solvation [13]. However, for any of these methods a detailed understanding of the chemistry of the unknown is required and, if these are not removed before the addition of HBQS dye, then there is a possibility of interaction between the dye and these

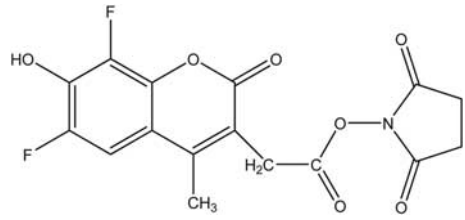

FIG. 4—*Chemical structure of marina blue.*

strong reactants. Since contaminated samples would contain an unknown impurity in small quantities, absorptive methods were examined as a preferred way to selectively remove the impurity, where these absorbents can also be removed effectively after the treatment. The absorbant chosen was activated carbon (or activated charcoal). The carbon is activated through treatment with oxygen to open up a large surface area between porous carbon structures [14], which gives it countless bonding sites to remove organic impurities. Activated charcoal is good at trapping organic impurities as well as some halides (e.g., chlorides); however, it has little affinity for metals and metal ions [15].

Marina Blue was chosen for this study as the spiked fluorescence impurity and different grades of activated charcoal as absorbents were tested. The Marina Blue was first dissolved in water and then added to 1 wt% ABF until the

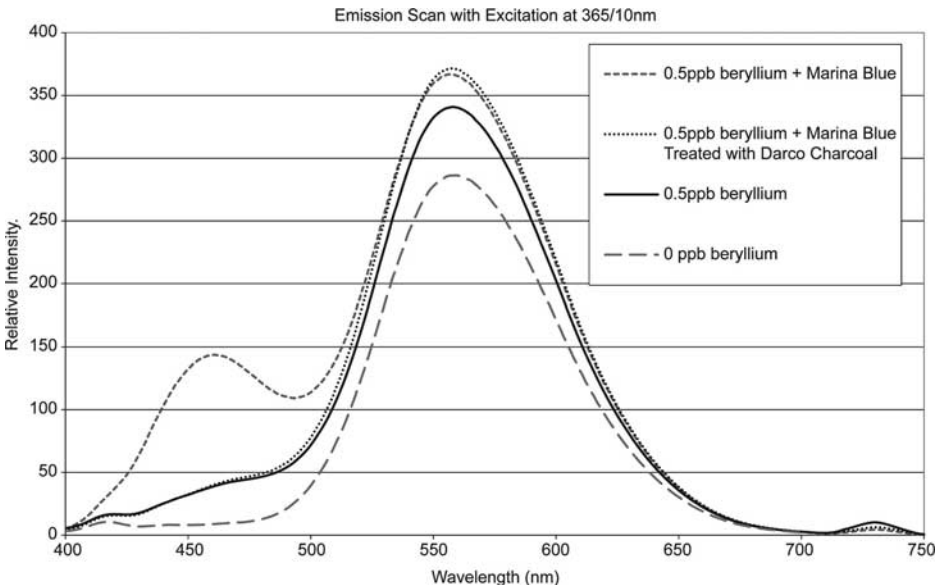

FIG. 5—*Emission scan showing the increase in fluorescence with addition of marina blue.*

fluorescence reading at 475nm was close to that of beryllium bound to HBQS in our reference standards (~1.34 ppb). The two different grades of activated carbon used were Darco G60, a steam activated carbon and a hydrochloric acid washed activated carbon.

Since activated carbon removes organic materials, it is important to add this to the dissolution solution and then remove the activated charcoal before the dye solution is added. In this way beryllium chelated to the HBQS dye is not removed. In some of the initial experiments the dye solution was added to the dissolution solution with activated carbon and the results were erratic. The preferred procedure, which is shown schematically in Fig. 6, prevents the removal of HBQS by the charcoal and maintains test sensitivity. The shaded boxes represent new steps added to the existing procedures (NIOSH analytical methods 7704 and 9110 and ASTM methods D7202 and D7458).

Typically the dissolution process for wipes and air filters uses 1 wt% ABF solution processed for 30 mins at 80 to 90°C. After the dissolution process, the solution was filtered through 0.2 μm hydrophilic polypropylene (GHP) filters. The concentration range of activated carbon tested was between 0.5 to 10 mg/ml in ABF solution, with the most favorable concentration found to be 1mg/ml. To test that the addition of activated carbon was not depleting or adding any beryllium (in case beryllium was present as an impurity in the activated carbon), the lowest concentration beryllium standard (0.5ppb) and one of the highest beryllium standards (40 ppb) were taken. These were then tested with and without the addition of activated carbon in 5 ml of dissolution solution and subjected to 30 mins at 80°C. The results are shown in the first part of Table 1. These results indicate that following the procedure in Fig. 6 there was no change in the beryllium content of the standards.

The bottom section of Table 1 lists experimental results for samples spiked with Marina Blue and then treated with activated carbon. Without the addition of activated carbon, the samples erroneously gave a high value for beryllium, whereas the activated carbon treatment completely eliminated the effect of Marina Blue, giving values for Be consistent with the standard value of 0.5 ppb.

The emission spectra for the data spiked with Marina Blue are shown in Fig. 7 and also confirm that the enhanced fluorescence signal due to Marine

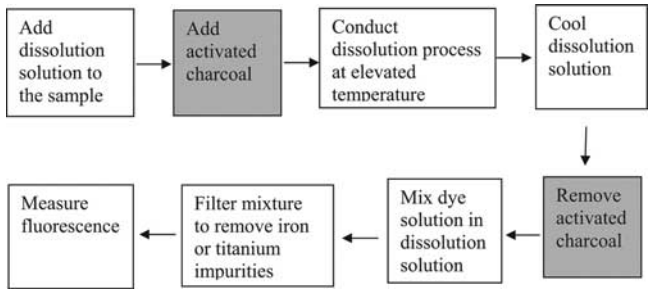

FIG. 6—*Schematic of preferred procedure to remove organic impurities.*

TABLE 1—*Addition of activated charcoal and Marina Blue to Be standards in the dissolution solution containing HBQS. Results are average of two readings.*

Sample (Be Standard Treated with Activated Charcoal at 1 mg/ml in 1 wt% ABF)	Actual [Be] Reading ppb[a]	Expected [Be] Reading ppb
Part 1: Effect of Activated Carbon Only		
0.5 ppb Be	0.50	0.50
0.5 ppb Be treated with Darco G-60 activated charcoal	0.52	0.50
0.5 ppb Be treated with activated charcoal washed with HCl	0.48	0.50
40.0 ppb Be treated with Darco G-60 activated charcoal	39.89	40.0
40.0 ppb Be treated with activated charcoal washed with HCl	40.12	40.0
Part 2: Effect of Activated Carbon on Samples Spiked with Marina Blue		
Marina Blue	1.34	0.00
0.5 ppb Be + Marina Blue	1.76	0.50
0.5 ppb Be + Marina Blue + DARCO G-60	0.53	0.50
0.5 ppb Be + Marina Blue + Carbon washed with HCl	0.49	0.50

[a] Based on standard beryllium calibration curve. Excitation/Emission 365/475 nm.

Blue is completely removed after treating with activated carbon. To confirm that the decrease in fluorescence signal is due to removal of Marina Blue and not thermal decomposition at the processing temperature in the dissolution solution, new samples were run where the fluorescence signal was read before and after the heat treatment. These experiments produced the same fluorescence value of 1.34, indicating that no quenching or thermal degradation of the Marina Blue dye was occurring.

1 wt% ABF solutions containing activated charcoal were tested for aging to investigate whether their efficacy changed if this mixture was stored and used over a period of time. Darco G-60 was dispersed at a concentration of 1mg/ml in the above solution and tested. This solution was stored at room temperature and periodically evaluated by taking an aliquot and adding Marina Blue spiked 0.5ppb Be standard solutions and analyzing them. After 105 days of storage, the stock solutions showed no degradation in their activity.

Addition of Activated Carbon During Soil Analysis for Beryllium

Recently the fluorescence method was shown to be very effective in determining beryllium content in soils [1,16] and has been approved as ASTM method D7458 [6]. However, some soils can have a high content of organic matter which can exhibit fluorescence and possibly interfere with the signal from HBQS. Further, for soil analysis the fluorescent method uses 3 wt% ABF heated to 90°C for 40 hs as the dissolution medium [1]. Thus, it was important to

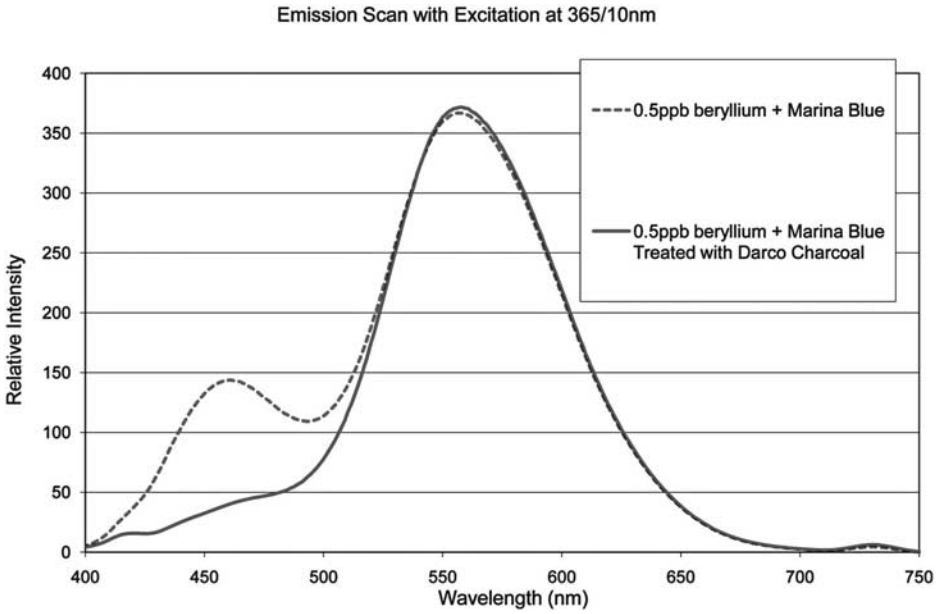

FIG. 7—*Emission spectra beryllium standard spiked with marina blue and then treated with DARCO G-60 activated carbon.*

establish if this dissolution protocol will be effective in removing organic contaminants from soils using activated carbon.

Under these dissolution conditions the addition of activated charcoal to Marina Blue spiked samples was tested and found to be as effective as in 1 %ABF solutions for 30 mins at 90°C. To test this, three NIST SRM soils were examined by the ASTM D7458 method where activated charcoal was added in the dissolution process. The SRM materials examined are listed in Table 2 and were chosen for their high organic carbon content (ascertained from the NIST certificate of analysis) which may comprise fluorescent impurities.

The soils were treated in 3 wt% ABF at 90°C for 40 hs with and without the addition of activated carbon and the results are listed in Table 2.

All the soil samples were largely unaffected by the addition of the activated carbon and yielded beryllium values consistent with the reference values, indicating they did not contain any appreciable interfering impurities and that the addition of activated charcoal did not affect the beryllium analysis.

Activated Carbon Process to Eliminate Background Fluorescence in Surface Sampling Wipes

Commonly used wipe materials were obtained and analyzed for beryllium background using fluorescence. Wipes were analyzed from different batches. The wipes were Ghost, Lead, Palintest wipes and Whatman 541 filters. The data are

TABLE 2—*Standard reference soils (SRM) and soil samples with activated carbon added.*

Sample	[Be] BeFinder[a] Method (ppb)	[Be] Reference Values (ppb)
SRM 2702 Marine Sediment without activated carbon	2.5	3
SRM 2702 Marine Sediment with Darco G-60 activated carbon	2.5	
SRM 1944 Waterway Sediment without activated carbon	1.8	1.6
SRM 1944 Waterway Sediment with activated carbon (HCl washed)	1.6	
SRM 2702 Marine Sediment without activated carbon	2.8	3
SRM 2702 Marine Sediment with activated carbon (HCl washed)	2.7	
SRM 2710 Montana Soil without activated carbon	2.5	2.5
SRM 2710 Montana Soil with activated carbon (HCl washed)	2.3	

[a] Peak transmission for Excitation/Emission for the filters used were 365/475 nm, respectively.

reported on two different lots of Palintest wipes as there was a visible difference between the two lots, Lot#110346 was yellowish in color and appeared as more stiff and dense paper towel, while Lot 26 011 was had no yellowish color and was softer and appeared to be a more porous structure. The analysis was done by treating the wipes in 20 ml of 1% ABF solution; the dissolution period was 60 mins at 85°C. The background fluorescence results on the wipes are shown in Table 3. This table also shows the size of each of the wipes, along with their moisture content. Other than the Whatman filters which are sold dry, the other wipe materials are individually packed in a wetted state.

The results demonstrate that Lead Dust wipes and Palintest wipes show elevated numbers for beryllium, particularly Lot#110346 for Palintest wipes. In order to see if this was being caused by organic fluorescent impurities in the wipes, these were analyzed by adding activated charcoal. In order to determine the amount of activated charcoal necessary, initial experiments were done with various levels of charcoal as shown in Table 4. These numbers were compared with a blank test, which included no wipes.

The results show an interesting trend: when 30 mg of activated charcoal was used in 20 ml of the dissolution solution, the beryllium numbers in the wipes decreased (compare these with no activated charcoal in Table 3), which showed that the elevated beryllium beryllium numbers in Table 3 were at least partly related to the fluorescent impurities. In addition at charcoal loading of 100 mg, the blanks started showing a beryllium number which increased at 200 mg. In all the other results, the beryllium numbers decreased at 100 mg but increased at 200 mg loading. Since activated charcoal is made by controlled combustion of vegetable matter, it is likely that they have a traceamount of beryllium contamination which is detected with increasing amount of activated

TABLE 3—*Beryllium analysis on various wipes by fluorescence and their characteristics.*

	Beryllium, μg ($n = 4$)		Water Content, g ($n = 4$)		Size,
	Average	Std. Deviation	Average	Std. Deviation	cm × cm (sq cm)
Blank (no wipe)	−0.006	0.001			
Ghost Wipe	–0.002	0.001	3.54	0.01	15 × 14 (208)
Lead Dust Wipe	0.013	0.001	1.84	0.05	20 × 13(250)
Palintest Wipe, Lot# 26011	0.007	0.000	1.80	0.04	18 × 13 (226)
Palintest Wipe, Lot#110346	0.059	0.001	1.98	0.01	18 × 13 (226)
Whatman Filter[a] 541	–0.004	0.001	0		4.7dia (17)

[a] This analysis reports the values on four Whatman filters that were analyzed together.

charcoal. This suspicion was confirmed by ICP-AES experiments on activated carbons, and the results are discussed below in Table 5. In addition, 30 mg of charcoal was not sufficient to remove all the fluorescent impurities across the board.

A proprietary process was developed at Berylliant to wash the activated charcoal before using it in the dissolution solution to remove the fluorescent impurities. This washing process was developed to eliminate the problem of recontamination of wipes. In order to test the efficacy of the washing process, samples of washed and as received (untreated) Darco 60 charcoal were analyzed for beryllium and iron. This analysis was done by treating these charcoals with 1% and 3% ABF solution for 1 h and 40 hs respectively at 90°C. The liquid phase was then analyzed by inductively coupled plasma-atomic emission spectroscopy (ICP-AES) to investigate if beryllium and iron could be extracted. The results are shown in Table 5. The limit of beryllium detection was < 0.0009 μg/ml of solution.

TABLE 4—*Effect of addition of activated charcoal in various amounts during dissolution.*

	Beryllium, μg After Treating with Darco 60 Activated Carbon ($n = 4$)					
	30 mg	Std. Dev.	100 mg	Std. Dev.	200 mg	Std. Dev.
Blank (no wipe)	−0.0028	0.0005	0.0040	0	0.0138	0.0005
Lead Dust Wipe	0.002	0	0.003	0	0.0128	0.0005
0 mg	−0.006	0.013	0.007	0.059		
Std. Dev.	0.001	0.001	0.000	0.001		
Palintest Wipe, Lot# 26011	0	0	0.0038	0.0005	0.0125	0.006
Palintest Wipe, Lot#110346	0.030	0.0022	0.007	0	0.0130	0

TABLE 5—*Measurement of extracted beryllium and iron using ammonium bifluoride aqueous solution (1 and 3% by wt/volume) from untreated and washed Darco 60.*

Sample	% ABF	Extraction Time (hr) at 90°C	Carbon (g)	ABF solution vol (ml)	Analysis		Extracted	
					Be μg/ml	Fe μg/ml	Be μg/g	Fe μg/g
Washed #1	3	40	0.192	10	< 0.0009	0.888	< 0.045	46.3
Washed #2	3	40	0.19	10	< 0.0009	1.068	< 0.045	56.2
								51.2[a]
Untreated #5	3	40	0.203	10	0.0016	11.665	0.079	574.6
Untreated #6	3	40	0.207	10	0.0016	11.785	0.077	569.3
							0.078[a]	572.0[a]
Washed #3	1	1	0.203	10				
Washed #4	1	1	0.195	10	< 0.0009	0.463	< 0.045	23.7
Untreated #8	1	1	0.209	10	0.0012	11.25	0.057	538.4
Untreated #9	1	1	0.195	10	0.001	10.16	0.051	520.9
							0.054[a]	529.6[a]

[a]Average of the above two values.

TABLE 6—*Effect of addition of washed activated charcoal Darco 60 at 200 mg/20 ml of dissolution solution.*

	Beryllium, μg ($n = 4$)					
	Results not Corrected for Water in Wipes		Results not Corrected for Water in Wipes		Results Corrected for Water in Wipes	
	No Added Beryllium	Std. Dev.	0.2 μg of Added Beryllium	Std. Dev.	0.2 μg of Added Beryllium	Std. Dev.
Blank (no wipe)	−0.0033	0.001	0.207[a]	0.0014	…	…
Ghost Wipe	−0.001	0	Not done			
Lead Dust Wipe	−0.0035	0.0006	0.175[a]	0.0014	0.191[a]	0.0015
Palintest Wipe, Lot# 26011	−0.0035	0.0006	0.192[a]	0.0007	0.209[a]	0.0008
Palintest Wipe, Lot#110346	−0.0025	0.001	0.17	0	0.187	0
Whatman 541	−0.004	0	Not done			

[a]Data on these samples are taken from two experiments ($n = 2$).

The results show that the washing process was effective in removing beryllium and iron, where the iron concentration was reduced by a factor of ten.

Table 6 shows the results for the various wipes when, during dissolution process, 200 mg of washed activated charcoal was added. The results are shown in three sets of columns, all using 200 mg of washed charcoal in 20 ml of dissolution solution. The first set of columns presents the data on wipes treated with washed charcoal, the second set of columns shows results when 0.2 μg of beryllium was added in the dissolution solution for each wipe, and the last set of columns is the same result as in the previous columns but corrected for the water content in the wipes.

The data in Table 6 show that using 200 mg of washed activated charcoal in 20 ml of dissolution solution was effective in removing the background fluorescence from all of these wipes, and did not interfere with the determination of beryllium present in the samples.

Conclusions

Recently standardized test methods have been developed to measure beryllium content by fluorescence. This investigation shows that in case where the samples are contaminated with organic fluorescent impurities that interfere with this measurement, one can effectively remove these impurities by using a simple procedure employing activated carbon black. Activated carbon was added to the dissolution solution that is used to dissolve beryllium particles from the media prior to fluorescence analysis. After the dissolution process, the activated carbon was removed and the beryllium specific fluorescent dye was added. Two types of activated carbon were tested and both were found to be highly effective in removing the organic impurity without compromising the beryllium measurement. This method was effective for all protocols used for wipes, filters and soils.

Washed activated carbon was not used during the dissolution process when the soils and Marina blue were analyzed; however, the levels of activated carbon required to remove fluorescence was low in those samples (1mg/ml of dissolution solution). Apparent beryllium contamination by activated carbon was only observed when higher level of activated carbon had to be used to douse all of the fluorescent impurities in the wipes, particularly Lot#110346 of Palintest wipes. The source contamination of trace beryllium in activated carbon was removable by washing. Since it is difficult to predict the contamination level of interfering fluorescent organic impurities, it is recommended that 10mg/ml of washed active carbon be used for all analyses where such impurities are suspected.

References

[1] Cronin, J. P., Agrawal, A., Adams, L., Tonazzi, J. C. L., MBrisson, M. J., White, K. T. Marlow, D. and Ashley, K. "Interlaboratory Evaluation of an Extraction and Fluorescence Method for the Determination of Trace Beryllium in Soils," *J. Environ. Monit*., Vol. 10, 2008, 955–960.

[2] Ashley, K., Agrawal, A., Cronin, J., Tonazzi, J. McCleskey, T. M. Burrell, A. K. and Ehler, D. S., "Ultra-Trace Determination of Beryllium in Occupational Hygiene

Samples by Ammonium Bifluoride Extraction and Fluorescence Detection Using Hydroxybenzoquinoline Sulfonate," *Anal. Chim. Acta*, Vol. 584, 2007, pp. 281–286.

[3] Agrawal, A., Cronin, J., Tonazzi, J., McCleskey, T. M., DEhler, D. S., Minogue, E. M., Whitney, G., Brink, C., Burrell, A. K., Warner, B., Goldcamp, M. J., Schlect, P. C., Sonthalia, P. and Ashley, K., "Validation of a Portable Fluorescence Method for the Measurement of Trace Beryllium in the Workplace Air and Wipe Samples," *J. Environ. Monit.*, Vol. 8, 2006, pp. 619–624.

[4] Ashley, K., McCleskey, T. M., Brisson, M., Goodyear, G., Cronin, J., and Agrawal, A., "Interlaboratory Evaluation of a Portable Fluorescence Method for the Measurement of Trace Beryllium in the Workplace," *Journal of ASTM International*, 2005, Vol. 2, No. 9.

[5] ASTM D7202-06, 2006, "Standard Test Method for Determination of Beryllium in the Workplace Using Field-Based Extraction and Fluorescence Detection," *ASTM International*, West Conshohocken, PA.

[6] ASTM D7458-08, (2008), "*Standard Test Method for Determination of Beryllium in Soil, Rock, Sediment, and Fly Ash Using Ammonium Bifluoride Extraction and Fluorescence Detection*," ASTM International, West Conshohocken, PA.

[7] *Beryllium in Air-by Field-Portable Fluorometry*, Analytical test method 7704, http://www.cdc.gov/niosh/docs/2003-154/pdfs/7704.pdf; *Beryllium in Surface Wipes-by Field-Portable Fluorometry*, Analytical test method 9110, http://www.cdc.gov/niosh/docs/2003-154/pdfs/9110.pdf, US National Institute for Occupational Safety and Health (NIOSH), Cincinnati, OH, NIOSH Manual of Analytical Methods (NMAM), 5th ed., 2007.

[8] *Beryllium and Compounds, as Be, Analytical test method 7102*, http://www.cdc.gov/niosh/docs/2003-154/pdfs/7102.pdf; *Elements by ICP (Nitric/Perchloric Acid Ashing)*, Analytical test Method7300, http://www.cdc.gov/niosh/docs/2003-154/pdfs/7300.pdf, US National Institute for Occupational Safety and Health (NIOSH), Cincinnati, OH, NIOSH Manual of Analytical Methods (NMAM), 4th ed., 2003.

[9] *Determination of Trace Elements in Waters and Wastes by Inductively Coupled Plasma-Mass Spectrometry*, Method 2008, US Environmental Protection Agency (EPA), https://www.nemi.gov/apex/f?p=237:38:5526697933954163::::P38_METHOD_ID:4665, Washington DC, 1994.

[10] Minogue, E. M., Burrell, A. K., McCleskey, T. M., and Taylor. T. P., "Development of a New Fluorescence Method for the Detection of Beryllium on Surfaces," *Journal of American Society for Testing Materials International*, Vol. 2, No. 10, 2006.

[11] "Alexa Fluor Dyes Spanning the Visible and Infrared Spectrum–Section 1.3," *Molecular Probes Handbook, A Guide to Fluorescent Probes and Labeling Technologies, 11th Edition,* Johnson, I., Spence, M.T.Z., Eds., Life Technologies Corporation (Invitrogen), Carlsbad, CA, 2010, http://www.invitrogen.com/site/us/en/home/References/Molecular-Probes-The-Handbook.html.

[12] Matsumiya, H., Hoshino, H., and Yotsuyanagai, T., *Analyst*, Vol. 126, 2002, pp. 2082.

[13] Armarego, W. L. F. and Chai, C. L. L., *Purification of Laboratory Chemicals*, 6th ed., Elsevier, New York, 2009.

[14] Marsh, H. and Reinoso, F. R., *Activated Carbon, Elsevier Science*, New York, 2005.

[15] Bandosz, T. J, *Activated Carbon Surfaces in Environmental Remediation and Technology*, 1st ed. Academic Press, New York, 2006.

[16] Agrawal, A., Cronin, J. P., Agrawal, A., Tonazzi, J. C. L., Adams, L., Ashley, K., Brisson, M., Brandy, D., Whitney, G., Burrell, A. K., McCleskey, T -M, Robbins, J, and White, K., "Extraction and Optical Fluorescence Method for the Measurement of Trace Beryllium in Soils," *Environ. Sci. Technol.*, Vol. 42, No. 6, 2008, pp. 2066–2071.

Reprinted from JAI, Vol. 8, No. 3
doi:10.1520/JAI103489
Available online at www.astm.org/JAI

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Beryllium Measurement in Commercially Available Wet Wipes

ABSTRACT: Analysis for beryllium by fluorescence is now an established method which is used in many government run laboratories and commercial facilities. This study investigates the use of this technique using commercially available wet wipes. The fluorescence method is widely documented [1,2] and has been approved as a standard test method by the ASTM International and the National Institute for Occupational Safety and Health. The procedure involves dissolution of samples in an aqueous ammonium bifluoride solution and then adding a small aliquot to a basic hydroxybenzoquinoline sulfonate fluorescent dye, (Berylliant™, Inc., Detection Solution Part #CH-2) and measuring the fluorescence. This method is specific to beryllium. This work will explore the use of three different commercial wipes spiked with beryllium as beryllium acetate or as beryllium oxide and subsequent analysis by optical fluorescence. The effect of possible interfering metals such as Fe, Ti, and Pu in the wipe medium is also examined.

Introduction

Sampling for beryllium is expected to increase both in the Department of Energy and Commercial Industry, as the health risk associated with berryllium exposure is more widely understood. In order to deal with this expected increase in samples, more efficient and less labor intensive methods were investigated.

Manuscript received October 14, 2010; accepted for publication February 3, 2011; published online March 2011.

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Cite as: Youmans-McDonald, L. D., Brisson, M., Bernard, M., Agrawal, A., Cronin, J. P. and Adams, L., "Beryllium Measurement in Commercially Available Wet Wipes," *J. ASTM Intl.*, Vol. 8, No. 3. doi:10.1520/JAI103489.

Current beryllium analysis by hydrofluoric acid (HF) or sulfuric acid (H_2SO_4) digestion [3] and the removal of spectral interferences by ion exchange [4] before measurement by sequential Inductively Coupled Plasma-Atomic Emission Spectrometer (ICP-ES) is very labor intensive, resulting in long turnaround times. One method that meets the requirements of low cost and fast turnaround is optical fluorescence [5], which has been recently established as a standard test procedure for determining beryllium in particles collected on wipe samples, air filters, and soil samples. These methods are available as ASTM D7202 [6] and ASTM D7458–08 [7] and NIOSH 7404 and 7102 [8]. Alternative methods used to analyze beryllium are graphite furnace atomic absorption spectrometry (GFAAS), inductively coupled plasma atomic emission spectrometry, and inductively couple plasma mass spectrometry (ICP-MS) [9,10]. Compared to the other methods, the fluorescence method is not sensitive to interference by other metals present [4,11] and has beryllium detection limits comparable to the most sensitive method using ICP-MS. Duffay and Archuleta [12] reported work on the collection efficiency of some of the wet and dry commercial wipes for analysis of beryllium. However, no work has been reported on the use of commercial individually packaged wet wipes as a host media for beryllium and their analysis by optical fluorescence; further, there is no established consensus standard specification for wet wipes that can be used for beryllium surface sampling. Commercially available wipes are not required to adhere to a standard that ensures their suitability for beryllium measurement. Hence, there is no requirement for any commercial provider that the wipes used for beryllium sampling will not vary in material, preservative, or even background beryllium concentration, and this is a concern for any analytical method used for beryllium measurement. Issues related to commercial wet wipes are variability in their composition, size, amount of wetting agent, and addition of additives such as surfactants and anti microbial agents and variations between lots. Specifically, when optical fluorescence is used as the analytical method, there is a possibility that in addition to any background from beryllium (if present), other constituents from the wipe may react or dissolve to give products which have an emission and excitation spectrum overlapping with the fluorescent spectrum of the dye associated with beryllium; this will manifest as a positive bias resulting in an increased beryllium concentration. In this study, three commercial wipes were investigated: Palintest™, Ghostwipe™, and Methyl Cellulose Ester (MCE) filters. The effect of these wipes on beryllium analysis was investigated by combining the commercial wipe with a beryllium oxide spiked MCE filter or by directly spiking the wipe with beryllium acetate and analyzing by optical fluorescence. Also added to the wipe during the ABF dissolution process where known quantities of metals such as Fe, Ti, and Pu to investigated whether these interactions will cause a systematic error.

Methods

Three types of wipe media were used in this study and their details are listed in Table 1. The Ghostwipes™ and the Palintest™ wipes were spiked with beryllium

acetate and the MCE filters with high fired BeO oxide (Standard reference Material 1877 from National Institute of Standards and Technology). To test the effect of interfering metals on the analysis, Fe (chloride), Ti (dioxide), and Pu were added to the mixture in concentrations of 10 mg, 2 mg, and 5×10^6 disintegations per minute (dpm). Two different Lot numbers of the Ghostwipes were tested to check for any variations between batches.

For determination of beryllium by fluorescence, the wipe samples were subjected to a dissolution process by placing them in 20 mL of 1% ammonium bifluoride (ABF) and heating to 90°C for 60 min. Because of the large size of the wipes, 20 mL of ABF was chosen to get complete emersion of the wipe so that beryllium could be effectively extracted in the dissolution solution. The liquid after extraction was then analyzed using 5× dilution process [1] where 0.4 mL of the dissolution solution was added to 1.6 mL of a detection solution containing 1.1m*M* HBQS (hydroxybenzoquinoline sulfonate), 1m*M* ethylenediamine tetraacetic acid, and 100 m*M* L-Lysine monochloride with the solution *p*H adjusted to 12.85 (Berylliant, Inc., Tuscon, AZ). The fluorescence measurements for beryllium quantification was performed using a Turner BioSystems Modulus fluorometer (Sunnyvale, CA) with a bandpass filter for excitation and emission. The filter transmission characteristics for excitation were 365 ± 10 nm and 480×5 nm, for excitation and emission. The fluorometer was calibrated using solutions of 0.1 ppb to 16 ppb of beryllium.

Results and Discussion

The unique aspect of the fluorescence method to detect beryllium is the use of the fluorescence dye (HBQS), which specifically binds to beryllium. The phenolic group binds strongly to beryllium where the six member ring has the ideal distance between O–O or N–O for chelating Be [11]. A tightly bound hydrogen bonded proton leads to a weak triplet emission at 580 nm for the dye solution. When the proton is displaced by a metal such as beryllium, peak fluorescence emission is observed between 475 to 480 nm [10].

Because of the large size of the commercial wipes, higher volumes of 1 wt% ABF were required (20 mL as opposed to 5 mL) to completely submerge the wipe during the dissolution process. This results in a higher dilution and requires a reevaluation of the detection limits of the method. To test the viability of the commercial wipes for use with the fluorescence method, their background fluorescence signal was first established. This was done for the Palintest™ and Ghostwipes™ spiked with beryllium acetate and MCE filters spiked with beryllium oxide. These data are shown in Table 2. The blank Palintest™ wipe has a significant background fluorescence signal higher than 0.02 μg whereas the Ghostwipe™ and MCE filter have background fluorescence signals within the method detection limits. The additive factor of this background signal is seen when the Palintest™ wipe is spiked with 0.04 μg of beryllium, as beryllium acetate. The recoveries for Palintest™ are all well above 100%. The Ghostwipes™ and MCE filters, on the other hand, have recovery values on average in range 93–103%. Because of the high background fluorescence of the

TABLE 1—*Commercial wet wipes and filters used for analysis of beryllium using optical fluorescence.*

Material	Properties	Manufacture
Palintest™ wipe	Meets ASTM E1792 [7] size 17.9 × 12.7 cm	Palintest USA, KY, USA
Ghostwipe™	Meets ASTM E1792 [7] size 15 × 15 cm (or 15 × 14 cm)	Environmental Express, SC, USA, www.envexp.com
MCE filter	Blank and spiked with beryllium acetate or oxide	High-Purity Standards, Charleston, SC, USA 29423

TABLE 2—*Background and Be spiked fluorescence data for commercial wipes.*

	Palintest™ Wipe (% recovery)	Ghostwipe™ (% recovery)	High-Purity™ MCE Filter (% recovery)
Blank: 0 μg Be[a]	0.06 μ g	< 0.005 μg	< 0.005 μg
	0.03 μg	< 0.005 μg	< 0.005 μg
	0.04 μg		< 0.005 μg
			< 0.005 μg
Spiked with Be acetate, 0.04 μg Be	0.061 μg (153 %)	0.04 μg (100 %)	
	0.057 μg (139 %)	0.041 μg (103 %)	
	0.064 μg (160 %)	0.04 μg (100 %)	
	0.049 μg (120 %)	0.037 μg (93 %)	
	0.063 μg (154 %)	0.038 μg (93 %)	
Spiked with Be Oxide, 0.004 μg Be			0.0038 μg (93 %)
			0.0038 μg (93 %)
			0.0038 μg (93 %)
			0.004 μg (100 %)
			0.004 μg (100 %)

[a]Filters were spiked with a concentration of beryllium that is less than the established method reporting limit. The quantification limit for MCE wipes was found to be 0.005 μg.

Palintest™ wipes, they were deemed unsuitable for use with optical fluorescence. Previous lots of Palintest wipes did not demonstrate this problem [2].

To establish the reproducibility of the low background fluorescence of the wet Ghostwipes™, two different Lots were tested as shown in Table 3, which shows no significant variation between the two Lots.

The reporting limit for MCE filters spiked with 0.08 μg of Be (as BeO) was determined by optical fluorescence, as shown in Table 4. The limit of detection (LOD) was calculated using standard statistical "t" test at 99 % confidence limits. The lower limit of quantification (LOQ) was obtained by multiplying LOD by 5 and then averaging the three values. The LOQ for beryllium on MCE Filter was, on average, 0.003 μg.

The method reporting limit for Ghostwipes™ is shown in Table 5 based on three wipe samples spiked with 0.10 μg of beryllium, as beryllium acetate.

TABLE 3—*Ghostwipe™ lot comparison for background fluorescence.*

Sample	Ave, ppb	Std Dev, ppb	Ave, μg	Std Dev, μg
Lot number Aug. 4	0.02000	0.00000	0.00800	0.00000
Lot number Oct. 29	0.02037	0.00111	0.00815	0.00044
Average	0.02022	0.00067	0.00809	0.00027

Aug. 4 data based on six samples, and three aliquots were measured.

Oct. 29 data was based on nine samples, and three aliquots were measured.

TABLE 4—*Method reporting limit for MCE filters.*

Standard conc. used =	0.08	0.08	0.08
Units	ppb	ppb	ppb
Other parameters/information:	Filter	Filter	Filter
Replicate number[1,2]	**Measured values**	**Measured values**	**Measured values**
1	0.070	0.070	0.050
2	0.070	0.070	0.070
3	0.080	0.080	0.070
4	0.080	0.070	0.080
5	0.080	0.070	0.040
6	0.090	0.070	0.050
7	0.080	0.080	0.060
8	0.070	0.080	0.070
9	0.090	0.070	0.080
10	0.080	0.080	0.060
[1,2]Number measurements (n) =	10	10	10
Standard deviation (S) =	0.007	0.005	0.013
[4]Standard conc./S =	11	15	6
[5]Student's t ($\sigma = 0.99, n - 1$) =	2.8214	2.8214	2.8214
[6]Lower limit of detection (LLD) =	0.02082	0.01457	0.03774
[6]Lower limit of quantitation (LLQ) =	0.10409	0.07285	0.18868
Lower limit of quantitation (μg) =	**0.002** μg/filter	**0.002** μg/filter	**0.004** μg/filter

TABLE 5—*Method reporting limit for Ghostwipes™*

Standard conc. used =	0.10	0.10	0.10
Units	ppb	ppb	ppb
Other parameters/information:	Ghostwipe	Ghostwipe	Ghostwipe
Replicate number[1,2...10]	**Measured values**	**Measured values**	**Measured values**
1	0.090	0.090	0.100
2	0.100	0.090	0.100
3	0.100	0.090	0.100
4	0.100	0.100	0.080
5	0.100	0.100	0.100
6	0.090	0.130	0.100
7	0.110	0.090	0.100
8	0.100	0.100	0.090
9	0.100		0.090
10	0.110		0.100
[1,2]Number measurements (n) =	10	8	10
Standard deviation (S) =	0.007	0.014	0.007
[4]Standard conc./S =	15	7	14
[5]Student's t ($\sigma = 0.99, n-1$) =	2.8214	2.9979	2.8214
[6]Lower limit of detection (LLD) =	0.01881	0.04066	0.01973
[6]Lower limit of quantitation (LLQ) =	0.09405	0.20329	0.09864
Lower limit of quantitation (μg) =	**0.009** μg/wipe	**0.020** μg/wipe	**0.010** μg/wipe

TABLE 6—*Recovery of Ghostwipes™ combined with High-Purity™ MCE BeO standards.*

Concentration	Expected Value, μg	Result, ppb	Result, μg	% Recovery
BeO 0.01[a]	0.01	0.14	0.014	140.0
BeO 0.01[a]	0.01	0.11	0.011	110.0
BeO 0.2	0.2	2.01	0.201	100.5
BeO 0.2	0.2	1.92	0.192	96.0
BeO 0.5	0.5	4.88	0.488	97.6
BeO 0.5	0.5	4.51	0.451	90.2
BeO 1.0	1	9.19	0.919	91.9
BeO 1.0	1	8.97	0.897	89.7

[a]Concentration below established method reporting limit.

Based on the three sets of experiments, the average LOQ for the Ghostwipes™ was 0.013 μg. In many beryllium contaminated environments, the beryllium present is in the oxide form. To determine the recoveries of BeO in the presence of Ghostwipes™, BeO spiked MCE filters were inserted in the dissolution tube along with blank Ghostwipe™ and 20 mL of ABF was added for dissolution at 90°C for one h. Although NIOSH methods 7704 and 9110 recommend a 30 min heating step, other studies [13] have shown that heating for one hour can give a better beryllium recovery (up to 20% improvement for BeO containing samples). The resulting solution was then analyzed as previously described. The recovery data for beryllium under such conditions are shown in Table 6.

At the low BeO concentration of 0.01 μg, the background from both the Ghostwipe™ and MCE filter are adding to the fluorescence signal, resulting in recovery values higher than 100%. As the value of the BeO increases above the established method reporting limit of 0.02 μg, the effect of the background signal is diminished and the recovery values are in the low to high 90% range.

Since many of the wipe samples will be taken in areas where other metals are present and even in some cases in the presence of radioactive materials, interference can be an issue. The metals, Fe and Ti, can cause yellow colored solutions, which can interfere with Be analysis results by producing a negative bias either via excessive absorption of the excitation beam or absorption of the emission signal by the solution. These metals were chosen as additional spike materials for the Ghostwipes™ in combination with the BeO spiked MCE filters. Also chosen was a low dosage of Pu. The recovery data for the interference testing are shown in Table 7.

The Ti and Pu additives show no interference effects and give recovery values in line with those shown on Table 6 where the additive materials were not present. The 10 mg Fe additive appears to affect the recovery in some instances. This stresses the importance of measuring spiked samples routinely with samples (in a similar way, ICP-ES requires interference correction for high iron levels). Even in the example shown, the average recovery was 98.7% with a standard deviation of 16.9%.

TABLE 7—*Interference testing, Fe/Ti/Pu in Ghostwipes™.*

Interference	Conc. Interference	Conc. Be, ppb	Be Result, ppb	Recovery	Mean (Standard Deviation)
Fe	10 mg	0.2	0.2	**100**	—
	10 mg	0.2	0.16	**75**	—
	10 mg	2	1.74	**87**	—
	10 mg	2	1.76	**88**	—
	10 mg	0.2	0.21	**105**	—
	10 mg	0.2	0.2	**100**	—
	10 mg	0.2	0.27	**135**	—
	10 mg	0.2	0.23	**115**	—
	10 mg	2	1.84	**92**	—
	10 mg	2	1.8	**90**	**99 (17)**
Ti	2 mg	0.2	0.21	**105**	—
	2 mg	0.2	0.22	**110**	—
	2 mg	2	1.72	**86**	—
	2 mg	2	1.69	**85**	**97 (13)**
Pu	$\sim 5 \times 10^6$ dpm[b]	0.2	0.21	**105**	—
	$\sim 5 \times 10^6$ dpm[b]	0.2	0.2	**100**	—
	$\sim 5 \times 10^6$ dpm[b]	2	1.85	**93**	**99 (6)**
HP-CRMD[a]	n/a	0.1	0.104	**104**	—
HP-CRME	n/a	0.2	0.201	**101**	—
Blank	n/a	0	0.04	**n/a**	—
Blank	n/a	0.01	0.04	**n/a**	—
Blank	n/a	0.01	0.02	**n/a**	—

[a]CRMD and CRME are spiked filters prepared by High-Purity with beryllium acetate in known concentrations indicated.
[b]Approximately 5×10^6 disintegrations per minute of alpha activity, contributed to plutonium.

Conclusions

Based on the information provided in this article, it is concluded that some commercially available wet wipes would be suitable for beryllium wipe measurement by fluorescence. Because there is no current standard that commercially available wet wipes must adhere to for beryllium measurement, wipes used would require lot testing to provide documentation of suitability.

Acknowledgments

The writers gratefully acknowledge Shirley Riley, Erika Lee, and Chad Durden for the generation of the data used in the tables provided. Mention of commercial products in this article does not imply endorsement by the writers, SRNS, SRS, or the U.S. Department of Energy (DOE).

References

[1] Ashley, K., Agrawal, A., Cronin, J., Tonazzi, J., McCleskey, T. M., Burrell, A. K., and Ehler, D. S., "Ultra-Trace Determination of Beryllium in Occupational Hygiene Samples by Ammonium Bifluoride Extraction and Fluorescence Detection Using Hydroxybenzoquinoline Sulfonate," *Anal. Chim. Acta*, Vol. 584, 2007, pp. 281–286.

[2] Agrawal, A., Cronin, J., Tonazzi, J., McCleskey, T. M., Ehler, D. S., Minogue, E. M., Whitney, G., Brink, C., Burrell, A. K., Warner, B., Goldcamp, M. J., Schlect, P. C., Sonthalia, P., and Ashley, K., "Validation of a Portable Fluorescence Method for the Measurement of Trace Beryllium in the Workplace Air and Wipe Samples," *J. Environ. Monit.*, Vol. 8, 2006, pp. 619–624.

[3] Ashley, K., and Oatts, T. J., 2009, "Sample Dissolution Reagents for Beryllium—Applications in Occupational and Environmental Hygiene," *Beryllium Environmental Analysis and Monitoring*, M. J. Brissonand A. Ekechukwu, Eds., Chap. 4, Royal Society of Chemistry (RSC), Cambridge, UK, pp. 89–101.

[4] Maxwell, S. L., III, Bernard, M. A., Nelson, M. R., and Youmans, L. D., "New Method for Removal of Spectral Interferences for Beryllium Assay using Inductively Coupled Plasma Atomic Emission Spectrometry," *Talanta*, Vol. 76, 2008, pp. 432–440.

[5] "Twice the Samples, Half the Time," *The Y12 Report No.*, Vol. 7(1), p. 7, Spring 2010, http://www.y12.doe.gov/news/report/toc.php?vn=7_1&xml=p07.

[6] ASTM D7202, 2005, "Standard Test Method for Determination of Beryllium in the Workplace Using Field-Based Extraction and Fluorescence Detection," *Annual Book of ASTM Standards*, Vol. 11.07, ASTM International, West Conshohocken, PA.

[7] ASTM D7458–08, 2008, "Standard Test Method for Determination of Beryllium in Soil, Rock, Sediment, and Fly Ash Using Ammonium Bifluoride Extraction and Fluorescence Detection," *Annual Book of ASTM Standards*, Vol. 04.09, ASTM International, West Conshohocken, PA.

[8] U.S. National Institute for Occupational Safety and Health (NIOSH), "Method Nos. 7704: BERYLLIUM in Air by Field-Portable Fluorometry., and Method Nos. 9110: BERYLLIUM in Surface Wipes by Field-Portable Fluorometry," *NIOSH Manual of Analytical Methods*, 5th ed., NIOSH, Cincinnati, OH, 2007.

[9] U.S. National Institute for Occupational Safety and Health (NIOSH), "Method Nos. 7102 and 7300," *NIOSH Manual of Analytical Methods*, 4th ed., NIOSH, Cincinnati, OH, 1994.

[10] U.S. Environmental Protection Agency (EPA), *Trace Elements in Water and Wastes—ICP-MS, Method No. 200.8*, EPA, Washington, D.C., 1994.

[11] Minogue, E. M., Burrell, A. K., McCleskey, T. M., and Taylor, T. P., "Development of a New Fluorescence Method for the Detection of Beryllium on Surfaces," *J. ASTM Int.*, Vol. 2, No. 10, 2006, Paper ID: JAI131689.

[12] Duffay, S. K., and Archuleta, M., "Comparison of Collection Efficiencies of Sampling Methods for Removable Beryllium Surface Contamination," *J. Environ. Monit.*, Vol. 8, 2006, pp. 630–633.

[13] Goldcamp, M. J., Goldcamp, D. M., Ashley, K., Ferback, J. E., Agrawal, A., Millson, M., Marlow, D., and Harrison, K., "Extraction of Beryllium from Refractory Beryllium Oxide with Dilute Ammonium Bifluoride and Determination by Fluorescence—A Multiparameter Performance Evaluation," *J. Occup. Environ. Hyg.*, Vol. 6, 2009, pp. 735–744.

Reprinted from JAI, Vol. 8, No. 10
doi:10.1520/JAI103487
Available online at www.astm.org/JAI

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The Feasibility of Studying the Health Implications of Surface Beryllium Contamination: A Review of Eight Industries

ABSTRACT: It has been suggested that beryllium can enter through either intact skin or breaks in the skin to initiate sensitization. Therefore, it is essential that all possible pathways of exposure be considered when assessing a work site for potential exposure to beryllium. A meta-analysis was done on the data from eight different industries that use beryllium or beryllium alloys, which were surveyed by Occupational Safety and Health Administration contractors, with reports being issued. During those visits, measurements were made to characterize worker exposure to skin and surface contamination levels of beryllium. Surface contamination evaluation followed the well-established protocol for lead surface contamination measurements using NIOSH Method 9100 and hand contamination using NIOSH Method 9102. It was found that both the arithmetic and geometric means of the surface samples were significantly higher in work areas as compared to administrative areas. Skin samples were also higher for individuals in production areas than for those in non-production areas. If skin contamination is a route of sensitization, these results would mean that past studies of beryllium exposure and subsequent disease might have been confounded by the lack of skin exposure data.

KEYWORDS: beryllium, skin contamination, industrial processes

Introduction

The measurement of one's exposure to total airborne beryllium dust alone might not be the best predictor of chronic beryllium disease (CBD) [1]. The particle size, surface area, number of particles, solubility, and chemical form of

Manuscript received October 20, 2010; accepted for publication August 8, 2011; published online September 2011.

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Cite as: McCawley, M., "The Feasibility of Studying the Health Implications of Surface Beryllium Contamination: A Review of Eight Industries," *J. ASTM Intl.*, Vol. 8, No. 10. doi:10.1520/JAI103487.

beryllium involved might all be relevant to the development of disease. It has also been suggested that beryllium can enter through either intact skin or breaks in the skin to initiate sensitization [2]. Recent studies have shown that particles less than 1 μm in diameter can penetrate intact skin that has been flexed [3]. Past epidemiology studies have not addressed this issue. Therefore, the case should be made that future studies should consider all possible pathways of exposure when assessing a work site for potential exposure to beryllium. One question that can be addressed now, which is pertinent to this issue, is whether, in a previous sample of industries using beryllium, the surface contamination level is sufficient to result in measurable skin exposure.

Background

Eight sites using beryllium or beryllium alloys were visited by Occupational Safety and Health Administration (OSHA) contractors who provided information on beryllium exposure and its control [4]. As part of the evaluation, both skin and surface sampling were done in each of the facilities. In summary, there were three machining plants, a metals recovery plant, a dental laboratory, two smelters, and a ceramics facility. A brief description of each operation follows.

Plant 1: Aluminum Beryllium Metal Machining Plant

The company was located in a 5000 ft^2 manufacturing facility and specialized in machining high tolerance aluminum, beryllium, aluminum-beryllium alloy, and other metals. The company operated three eight-hour shifts per day, five to six days per week. The facility had 11 full-time employees. Eighty percent of the machining was performed on aluminum, 19.9% on aluminum beryllium, and 0.1% on beryllium metal.

Plant 2: Beryllium Recovery Plant

This recycling facility is recognized for its metals recovery capability in the automotive, electronic, jewelry, and metal coating and fabrication industries. Copper beryllium's unique properties make it a material of choice for certain specialized electronics applications. Due to their high strength, high conductivity, and resistance to elevated temperatures, electronic component manufacturers make extensive use of copper beryllium alloys in connectors for automotive, computer, telecommunications, and information transmission equipment. Copper beryllium alloys are often present in the input products and specialized electronics systems that are introduced into metal recovery operations at this facility.

The company was located on a 160-year-old site that originally operated as a sawmill and textile mill in 1841. The site, approximately 48 acres with a total size including all buildings of approximately 110 000 ft^2, has operated as a precious metals recycling facility since 1973. The company specializes in the business of buying and processing materials that contain precious metals (gold, silver, platinum, palladium, ruthenium, rhodium, and iridium) and selling the

metals recovered from these materials. Secondary materials containing precious metals are received from a variety of industries and are assayed and prepared for shipment to an appropriate off-site smelter. The facility process capabilities are characterized as follows: material receipt and handling, mechanical preparation, granulation/shredding, thermal reduction, ball milling, screening, blending, melting, drying/grinding, and electrowinning.

The company operated primarily with one eight-hour shift per day, five days per week, 50 weeks per year. The company employs approximately 110 personnel, of which 75 are employed at the location visited, with an estimated 18 full-time workers reportedly having a potential for beryllium exposure. The facility processes approximately 15 000 000 pounds of electronics scrap per year and was reportedly at 50 % of capacity at the time of the visit.

Plant 3: Beryllium Ceramics Plant

The company was originally founded in 1956 as a beryllium oxide manufacturer. The company was a pioneer in the research and development of high purity oxide ceramics. In the early 1960s it developed the technology to press, extrude, and fire BeO in both standard and custom shapes to suit a wide variety of applications. In the late 1960s and early 1970s, precision ceramic grinding machinery was installed, along with the technology and equipment needed for drilling, metallizing, lapping, polishing, and dicing operations. During the period of peak production, the company operated with a schedule involing three eight-hour shifts and employed over 500 workers.

Plant 4: Beryllium and Aluminum Beryllium Metal Alloy Machining and Fabrication Plant

The company was located in a 100 000 ft^2 manufacturing facility and specialized in high tolerance beryllium metal and aluminum beryllium machining. The company operated two eight-hour shifts per day, five to six days per week. The facility had 214 employees, of whom 110 worked in the production area of the machine shop, with 71 on the day shift and 39 on the evening shift. Approximately half of the production area workers machined beryllium at least part of the time. However, the office workers also, on occasion, visited the beryllium work area. Because all employees had access to the production area, all employees were considered as beryllium exposed.

The company had over a dozen lathes and two dozen computer controlled mills, all of which could be used to process beryllium-aluminum alloy, beryllium, or aluminum. Some of the milling machines were enclosed, but all had ventilation supplied. Lapping, grinding, deburring, plating, heat treating, and manual milling were also performed as needed, along with a submerged electrical wire discharge cutting process called electric discharge machining. The pieces worked on ranged in size from a centimeter to several meters in length. The components produced were largely for defense and aerospace applications. Multiple pieces might be produced hourly, but some pieces require months or even years for completion, depending on their complexity and size. There is no standard

production flow, with work going from one machine to another. Some pieces could be produced using a single machine, whereas others might require several processing steps on multiple machines. In addition to the machining, there was an optics department that specialized in making high tolerance mirrors, primarily with nickel, but on a beryllium substrate, although beryllium parts were also lapped there. There was a gas bearings department that also used beryllium.

Plant 5: Dental Laboratory

The laboratory was located in a 300 ft^2 suite in the basement of a 35-year-old building in a health sciences school. The lab operated one eight-hour shift per day, five days per week. The facility had one employee on the day shift.

There were three rooms joined together in this laboratory. The primary work location was the grinding lab. A bench with a spinning wheel was located there for the technician to operate. Most of the technician's time was spent in this room. Next to this room was the casting room where the metal was melted and the casting was done. This room could be accessed only through the common lab area, in which no beryllium work was done. Several other technicians worked in this common room from time to time. There was no physical separation between the rooms, and they shared the same ventilation system.

Plant 6: Copper Beryllium Alloy Stamping Plant

The company was located in an approximately 20 000 ft^2 manufacturing facility and specialized in the precision stamping, forming, and plating of copper beryllium parts. The company operated one nine-hour shift per day, four and a half days per week. The facility had 22 employees. Copper beryllium was not used during every shift for any of the machines, and might be used for only part of any given shift.

Materials were generally received as plates or strips of beryllium copper. The alloys used were numbers 25, 10, 17410, and 7717. Material was stored onsite until required. The operators retrieved the raw material from storage and loaded the machines with the appropriate dimensions and material specifications. "Stamping" is a term used to refer to various press forming operations including coining, embossing, blanking, and pressing. The operations most commonly done were blanking, piercing, forming, and drawing. These operations were done with dedicated tooling, also known as hard tooling. Hard tooling is used to make high volume parts of one configuration. In the production process, a die is selected depending on the pattern required, and the material can be placed into either the stamping machine or the forming machine, or sometimes into both machines. The stamping machine is used to cut out patterns, much as a cookie cutter would. The forming machine is used to put bends and depressions into stamped parts. Workers sat at the machines, manually controlling the process and handling parts as they were produced. Manufactured parts coming from the machines were then placed into containers. These parts could be heat-treated, cleaned, dried, and plated. The assembly of parts could be done if required, and quality control was performed. Finished parts were then packaged and shipped to the customer.

Plant 7: Copper Beryllium Casting Plant

The company's core expertise was melting and casting beryllium copper and other beryllium containing alloys; over 1×10^6 pounds were melted and casted annually. Inert gas cover and degassing technology were utilized for melting and casting operations. Automatic furnace controls and ingot mould conveyers produced standard five-pound and two-pound ingot configurations. All beryllium alloys were manufactured utilizing either pure metallic beryllium or certified beryllium copper master alloy. No recovered, recycled, or purchased scrap was utilized in the standard production processes.

The company produced BeCu, BeNi, and BeAl casting and master alloy ingot; BeCu semi-continuous as-cast 21 in. diameter input billets; BeCu forged and turned precision input billets; BeCu forged rod and plate products; and BeCu large diameter hollows and custom forged products. The fractional content of beryllium in the products varied from 0.35 % to 10.5 % in the master alloy.

Administrative work was done on the day shift. Casting was done during the night shift, with only one casting done on an average night. Three people worked the night shift. Copper ingots and copper beryllium master alloy ingots were loaded into the pot before melting began. The ingots were mostly unloaded by hand into the pot from crates hoisted into place above the melting pot by the forklift. All three workers were involved in removing the approximately 12 in. long ingots from the crates and filling up the furnace.

Once the furnace was loaded, heat was applied and the melt began. Dross floats to the surface during the melt and needs to be skimmed off. Also, the melt needs to be sparged in order to ensure complete mixing (sparging involves the introduction of gas into the furnace to stir the melt). The majority of the shift, however, was spent waiting for the melting to be completed.

When the melt was complete, the pour began. The furnace was tilted, in-place, to pour the molten metal into moulds. The type of mould selected depended on the customers' specifications and could change from pour to pour. A number of moulds were available, as noted above in the list of products. The cast materials were allowed to cool and were then placed into containers for shipment, by either hand or forklift. Larger moulds could be trimmed using a band saw before being shipped. The band saw could have a lubricant stream that was used during cutting.

Plant 8: Non-ferrous Forging and Machining Plant

The company was a manufacturer and distributor of forged copper, including beryllium copper, chrome copper, and aluminum bronze in plates, blocks, bars, or rings, and other copper alloy forgings for plastic mould tooling, resistance welding applications, metal melting liners, end caps, and bearing components. Metal conversion services were also provided to customers who supplied their own metal materials. Services included the recommendation of alloys, properties, and designs for tooling applications; concurrent design; fabrication; heat treating to meet various temper and grain size requirements; near net shape processing; rough or finish machining; testing; and technical certifications.

The company had over 50 000 ft^2 of manufacturing space, and the operating schedule involved a single ten-hour shift, four days per week, with a total of 40 workers employed. There were 25 employees in the production shop, with 5 having direct involvement with CuBe alloys.

Methods

Surface Samples

Surface contamination evaluation followed the well-established protocol for lead surface contamination measurements, as did skin sampling. The exact surface locations were randomly selected to reflect all types of areas with which a worker might come in contact. These survey data were meant to answer the question of what the range of concentrations might be, rather than what specific processes and operations contributed to each sample. Surface sampling was done by marking off a 100 cm^2 area, using a plastic template with a square hole 10 cm on each side that was placed on the surface, with the corners marked with a moistened towelette. The template was then removed and the perimeter lined out with the same moistened towelette. The inside of the square was then wiped according to NIOSH Method 9100 [5], and the towelette was placed in a screw-top glass vial for analysis. The template was cleaned before reuse. Workers' gloves were also wiped, but at a separate time from the hand wiping so as not to disrupt the ordinary wear of the glove immediately before skin sampling.

Hand Wipe Samples

Hand wipe samples taken according to NIOSH Method 9102 [5] were obtained by asking study participants to wipe their hands before the end of their shift or during the shift, at least two hours after their last hand washing. They were instructed to lift a fresh wet wipe from an open container and to thoroughly wipe both hands (including the front and the back, up to the wrists, and each finger), removing as much visible dirt as possible. The wipe was then placed in a labeled screw-top glass vial. The hand wiping exercise was supervised and timed for 30 s by the investigators in order to ensure consistency from subject to subject. A pen tracing on graph paper with a millimeter scale as far as the wrist, where the wiping stopped, was taken of the subject's right hand in order to estimate the total surface area of the participants' hands. The number of 1 mm blocks on the graph paper within the tracing was counted and multiplied by four to give the total surface area of both hands. The concentration of beryllium on the workers' hands is reported in micrograms of beryllium per 100 cm^2 of estimated hand surface area.

Laboratory Analysis

OSHA's Salt Lake City Laboratory performed the laboratory analysis. Gravimetric results were reported to the nearest microgram. The beryllium mass was determined for surface wipes and hand wipes using NIOSH Method 7102 [5],

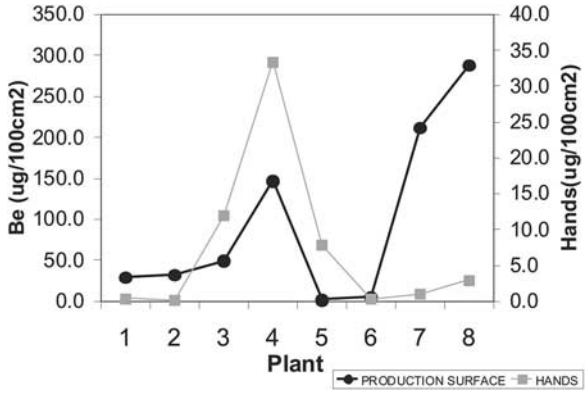

FIG. 1—*Average beryllium concentration on surfaces in production areas and on workers' hands.*

graphite furnace atomic absorption spectrophotometry. Field blanks made up 10% of the samples analyzed, and another 5% were media blanks.

Results

It appears from Fig. 1 that hand contamination follows a general trend with surface contamination in each plant, although not with quite the same proportion in each plant. With an approximately 300 cm^2 area on each worker's hand, there could be as much as 100 μg of beryllium, on average, on each of the hands of the workers in Plant 4. This figure would seem to indicate that surface contamination might generally be an indicator of the risk of dermal exposure but only a rough approximation of dermal exposure, and that direct measures of dermal exposure might be necessary for epidemiologic studies. Table 1 demonstrates that the surface contamination is usually higher in production areas than in non-production areas, which should not be surprising. The difference between many of the plants is by orders of magnitude. The geometric mean was found by taking the average of the log of the concentration values, averaging the log values, and then taking the antilog of that mean. The geometric mean tends to lessen the effect of a small number of extreme values. In Table 2 the large difference between the geometric and arithmetic means for the surface samples indicates that a broad range of contaminated surfaces were seen in each of the plants.

Discussion

In plant 1 there were five major machining centers, which together contained four computer numerically controlled milling machines and one lathe, all of which were enclosed, automated machining equipment. All equipment was operated by machinists, three of whom were evaluated during this site visit. Additionally, after machining, the workers performed deburring on certain products as a manual process for which specialized controls were used. Of all the plants in this study, only in plant 1 was there a glove policy in place

TABLE 1—*Individual plant results for beryllium contamination.*

	Arithmetic Mean, $\mu g/100\ cm^2$	Standard Deviation, $\mu g/100\ cm^2$	Number of Samples
Plant 1			
Surface production area	29.5	75.1	16
Surface non-production areas	1.5	2.0	9
Under gloves production area[a]	0.5	0.4	7
Plant 2			
Surface production area	32.7	128.0	29
Surface non-production areas	0.1	0.1	6
Hands production area	0.2	0.3	25
Hands non-production areas	<0.1	<0.1	3
Plant 3			
Surface production area	49.6	84.8	22
Surface non-production areas	0.5	0.6	7
Hands production area	11.9	20.5	12
Hands non-production areas	2.3	2.4	2
Plant 4			
Surface production area	147.2	564.8	88
Surface non-production areas	2.4	3.4	18
Hands production area	33.5	44.3	6
Plant 5			
Surface production area	2.0	4.1	18
Hands production area	7.8	-	1
Plant 6			
Surface production area	5.4	14.3	34
Surface non-production areas	<0.1	-	3
Hands production area	0.3	0.5	11
Plant 7			
Surface production area	212.0	185.3	13
Surface non-production areas	110.2	244.5	12
Hands production area	1.0	1.5	6
Plant 8			
Surface production area	288.7	1022.1	21
Surface non-production areas	0.4	0.5	4
Hands production area	3.0	4.7	19

[a] These samples were taken from hands after gloves were removed, in an area where workers wore gloves.

TABLE 2—*Comparison of arithmetic and geometric mean surface level beryllium contamination.*

	Arithmetic Mean, $\mu g/100\ cm^2$	Geometric Mean, $\mu g/100\ cm^2$
Plant 1		
Surface production area	29.5	3.9
Surface non-production areas	1.5	0.6
Plant 2		
Surface production area	32.7	2.3
Surface non-production areas	0.1	(<0.1)
Plant 3		
Surface production area	49.6	21.8
Surface non-production areas	0.5	0.3
Plant 4		
Surface production area	147.2	3.3
Surface non-production areas	2.4	0.2
Plant 5		
Surface production area	2.0	0.9
Plant 6		
Surface production area	5.4	0.7
Surface non-production areas	<0.1	<0.1
Plant 7		
Surface production area	212.0	94.4
Surface non-production areas	110.2	22.3
Plant 8		
Surface production area	288.7	17.0
Surface non-production areas	0.4	0.2

requiring the strict use of gloves. As can be seen from the data, this might be part of the reason that exposures and the potential for exposures might have been lower here than for most other plants. Enclosures around the equipment almost certainly contributed to the lower levels of surface contaminations as well. However, this is still somewhat speculative, because the objective of the reports cited was to simply document what existed, not what caused it to exist.

In plant 2 there were four operations at the site having a potential for beryllium exposure: mill/blending, thermal reduction, melting, and shredding mill. It is important to note that workers in the aforementioned processes and operations have potential exposures to lead and cadmium. The facility has characterized exposures to lead and cadmium and has implemented controls, personal protective equipment, and work practice control requirements in accordance with applicable regulatory standards. The lower levels of beryllium surface

contamination and dermal exposure might be due to the already heightened awareness of the potential for toxic exposures to other materials.

At the time of the survey, plant 3 was operating at an estimated 10% of capacity but had over 35 000 ft^2 of manufacturing space. It operated with a single eight-hour shift and had 14 workers employed. The low production levels, more than any exceptional controls, were probably responsible for the less than maximal surface contamination relative to the other plants.

Plant 4 had a history of diagnosed beryllium disease in its workers. There was no distinct separation between production and office areas, allowing employees to come and go between the areas, possibly spreading contamination. Although some control measures had been taken, other places where controls should have been in place showed a lack of adequate control. This might be why the plant had one of the highest levels of contamination.

Plant 5 had only one employee working on a low number of very small objects containing beryllium. Controls were virtually absent. The low production was the probable reason that this plant had lower contamination levels.

The alloy stamping plant (plant 6) had the lowest contamination levels and was the least energetic of those observed (it had the lowest air contamination level of any of the production areas, as well). Controls directed at reducing beryllium exposure were absent. However, copper beryllium was not used on every shift for any of the machines and was usually used for only a part of any given shift. The lower surface and dermal contamination is probably due to those latter effects rather than to the engineering design of the process.

Plants 7 and 8 were operated as metal refineries without reference to the toxicity of beryllium. Controls were minimal. It is presumed that the higher surface contamination was reflective of this.

Epidemiological studies to date have neglected or dismissed the possible effects of skin exposure, either drawing conclusions without measuring [6] or not including the analysis [7] of the skin exposure when measured (3.8 versus 1.1 $\mu g/100\ cm^2$ production versus nonproduction areas) in plants with known disease. The current American Conference of Governmental Industrial Hygienists threshold limit value for beryllium addressed only air exposure because there are no studies with data on skin exposure to reference. What needs to be addressed is the potential confounding effect of skin exposure, which could lead to sensitization and, with subsequent lung burden (from air exposure), then lead to lung disease.

In order to consider the possibility of establishing an epidemiologic investigation of the relationship between skin exposure and sensitization, it is necessary to have certain conditions. It is proposed that these conditions are as follows:

1. A source of exposure, which results in.
2. A quantifiable level of exposure across.
3. A range of exposures.
4. The range of exposures must include a putative level thought to cause an effect—in this case, sensitization.

It is obvious in this cross-section of plants performing varying kinds of work on beryllium and its alloys that there is a measurable source of exposure

to the skin on the hands throughout the production areas of the plant, and often in non-production areas, thereby meeting criterion 1 above. Figure 1, which shows a similar trend between hand contamination levels and surface contamination levels, indicates that surface contamination is the likely source. The levels found by OSHA were reported with a limit of quantification of 0.1 μg. This is in agreement with recent work [8] that shows a limit of detection for surface samples of 0.05 μg. The sample of plants for the OSHA reports therefore also meets criterion 2 above.

There is a distinct difference, for the most part, for both surface and hand concentrations between the production and non-production areas, with the production areas showing significantly higher contamination. This, then, achieves the first three proposed criteria for considering an epidemiologic study of skin exposure. The fourth criterion is the most problematic, in that no one has yet proposed a level that is either proven safe or hypothesized to cause sensitization. Unfortunately, most of the plants in this report did not screen for disease, and so disease rates are not available. One recent study [9], however, notes that there is a significant difference between sensitization rates in areas of a beryllium alloy plant between production and office areas, and the levels of airborne beryllium were generally lower than any published estimate of the exposure capable of causing sensitization or disease (geometric mean = 0.003 $\mu g/m^3$ and maximum = 0.02 $\mu g/m^3$). The same paper conducted a surface sampling analysis and found that surface contamination levels (also significantly different) were 0.95 and 0.05 $\mu g/100\ cm^2$ for production and office areas, respectively. If it is assumed that the production area contamination level of 0.95 $\mu g/100\ cm^2$ is the cause of the sensitization, then all of the plants in the OSHA survey have sufficient levels of contamination to cause sensitization. The fourth criterion is therefore met.

The plants in the OSHA reports were informed that the measurements would be given to OSHA, but with their names removed. One might assume that these plants, in choosing to cooperate, probably represent the best of the controlled plants. Becaus plants that did not wish to cooperate were not sampled, it is impossible to say for sure what relative level of surface cleanliness and control the plants studied actually represent. However, as the levels of surface contamination and dermal exposure that did occur in the plants studied are sufficient to warrant inclusion in an epidemiological study, and given that the assumption that these plants are the best controlled is not unreasonable, an epidemiological study of the relationship between dermal exposure and beryllium sensitization seems to be clearly warranted.

Conclusion

A biologically plausible hypothesis has previously been proposed relating skin exposure to beryllium sensitization [3]. Examining the data collected for OSHA in order to evaluate the current levels of exposure in various industries using beryllium, it is clear that dermal exposures are sufficient ($>0.95\ \mu g/100\ cm^2$) to warrant a study to prove or disprove the skin hypothesis, and that sampling and analytical methods are available with sufficient sensitivity to support that study.

Because sensitization seems to be an important step in the development of CBD and because the skin is a plausible means of exposure [10], unless the skin hypothesis can be dismissed, an airborne exposure limit might be difficult to support on its own.

References

[1] McCawley, M. A., "Rationale for Sampling Deposited Submicrometer Beryllium Particulate Matter," *J. Occup. Environ. Hyg.*, Vol. 6, 2009, pp. 789–793.

[2] Day, G. A., Stefaniak, A. B., Weston, A., and Tinkle, S. S., "Beryllium Exposure: Dermal and Immunological Considerations," *Int. Arch. Occup. Environ. Health*, Vol. 79, 2006, pp. 161–164.

[3] Tinkle, S. S., Antonini, J. M., Rich, B. A., Roberts, J. R., Salmen, R., DePree, K., and Adkins, E. A., "Skin as a Route of Exposure and Sensitisation in Chronic Beryllium Disease," *Environ. Health Perspect.*, Vol. 111, 2003, pp. 1202–1208.

[4] Eastern Research Group Inc., "Site Visit Report," *Report Nos. 1–9, Task Order 41, Contract No. J-9-F-9-0010*, Office of Regulatory Analysis, Directorate for Evaluation and Analysis, Occupational Safety and Health Administration, Washington, DC, 2003–2004.

[5] *NIOSH Manual of Analytical Methods*, 4th ed., Schlecht, P. C., and O'Connor, P. F., Eds., National Institute for Occupational Safety and Health, Washington, D.C., 1994.

[6] Rosenman, K., Hertzberg, V., Rice, C., Reilly, M. J., Aronchick, J., Parker, J. E., Regovich, J., and Rossman, M., "Chronic Beryllium Disease and Sensitization at a Beryllium Processing Facility," *Environ. Health Perspect.*, Vol. 113, 2005, pp. 1366–1372.

[7] Schuler, C. R., Kent, M. S., Deubner, D. C., Berakis, M. T., McCawley, M., Henneberger, P. K., Rossman, M. D., and Kreiss, K., "Process-Related Risk of Beryllium Sensitization and Disease in a Copper-Beryllium Alloy Facility," *Am. J. Ind. Med.*, Vol. 47, 2005, pp. 195–205.

[8] Dufresne, A., Turcotte, V., Golshahi, H., Viau, S., Perrault, G., and Dion, C., "Solvent Removal of Beryllium from Surfaces of Equipment Made of Beryllium Copper," *Ann. Occup. Hyg.*, Vol. 53, 2009, pp. 353–362.

[9] Day, G. A., Dufresne, A., Stefaniak, A. B., Schuler, C. R., Stanton, M. L., Miller, W. E., Kent, M. S., Deubner, D. C., Kreiss, K., and Hoover, M. D., "Exposure Pathway Assessment at a Copper-Beryllium Alloy Facility," *Ann. Occup. Hyg.*, Vol. 51, 2007, pp. 67–80.

[10] Bailey, R. L., Thomas, C. A., Deubner, D. C., Kent, M. S., Kreiss, K., and Schuler, C. R., "Evaluation of a Preventive Program to Reduce Sensitization at a Beryllium Metal, Oxide, and Alloy Production Plant," *J. Occup. Environ. Med.*, Vol. 52, 2010, pp. 505–512.

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Reprinted from JAI, Vol. 8, No. 5
doi:10.1520/JAI103477
Available online at www.astm.org/JAI

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Evaluation of Asbestos in Dust on Surfaces by Micro-Vacuum and Wipe Sampling

ABSTRACT: Micro-vacuum and wipe sampling methods are routinely used to measure surface dust contamination in both occupational and environmental settings. Both methods are subject to variability in their efficiency of dust collection resulting primarily from textural surface characteristics and dust loading. As part of a study to determine contamination levels of asbestos and other World Trade Center related contaminants in the wall cavity of a high-rise office building, paired samples were collected from 15 locations. The surface type of 14 locations was concrete-masonry block; the remaining surface type was a polyvinyl chloride coating. Micro-vacuum wipe pairs were obtained from the same wall component at contiguous locations. A template was used to ensure that equal 100-cm^2 areas were consistently sampled. Micro-vacuum and wipe samples were collected and analyzed for asbestos using ASTM Methods D5756 and D6480, respectively. The average surface concentration reported by the micro-vacuum samples was numerically higher than the wipe samples, but the difference was not statistically significant ($P = 0.195$). Both methods yielded an equal number of samples below the analytical sensitivity; the false-negative rates were the same for each method. Micro-vacuum and wipe sample concentrations were not correlated ($R^2 = 0.207$). The length and width of asbestos structures collected by wipe samples was significantly larger than by micro-vacuum samples ($P = <0.001$). Micro-vacuum sampling for asbestos in dust on rough surfaces yields numerically higher concentrations of asbestos. Despite variations in the relative efficiency of particle collection by these methods, the data pairs

Manuscript received October 12, 2010; accepted for publication April 5, 2011; published online May 2011.

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Cite as: Kominsky, J. R. and Millette, J. R., "Evaluation of Asbestos in Dust on Surfaces by Micro-Vacuum and Wipe Sampling," *J. ASTM Intl.*, Vol. 8, No. 5. doi:10.1520/JAI103477.

demonstrate that micro-vacuum and wipe samples produced statistically equivalent results, with micro-vacuum samples being more efficient on the rough surface tested.

KEYWORDS: asbestos, settled dust, surface sampling, wipe, micro-vacuum

Introduction

Interest in assessing asbestos in surface dust can be traced back at least to 1935 where, to evaluate occupational hygiene conditions in six asbestos plants, dust that had settled on rafters was collected and analyzed for asbestos by polarized light microscopy [1]. The earliest use of a micro-vacuum sampling device to collect asbestos from surfaces appears to be that reported in 1970 to assess dust generated from machining of amosite- and chrysotile-containing materials [2]. Millette and Hays (1994) reviewed the history of settled dust sampling methods [3]. Several studies have noted advantages and disadvantages of surface sampling methods for asbestos in settled dust [4–6].

Although a direct relationship to exposure risk and asbestos surface contamination is equivocal, asbestos surface dust sampling is used as a tool for contamination and exposure assessments [7]. The measurement of asbestos in settled dust is important because asbestos may become suspended in air and contribute to airborne exposure [8]. The primary purpose of surface sampling is to determine the loading of asbestos in dust on surfaces, which represents both the current and historical asbestos accumulation.

Three methods for measuring asbestos in settled dust have become ASTM standards through the efforts of Subcommittee D22.07 on *Sampling and Analysis of Asbestos*. These methods are: D5755-03, *Test Method for Micro-Vacuum Sampling and Indirect Analysis of Dust by Transmission Electron Microscopy for Asbestos Structure Number Surface Loading*; D5756-02 (2008), *Test Method for Micro-Vacuum Sampling and Indirect Analysis of Dust by Transmission Electron Microscopy for Asbestos Mass Loading; and* D6480-05, *Test Method for Wipe Sampling of Surfaces, Indirect Preparation, and Analysis for Asbestos Structure Number Loading by Transmission Electron Microscopy*.

These methods collect surface dust by vacuuming [D5755 and D5756] or wiping [D6480] a known surface area and use an indirect technique of sample preparation to disperse the particles. In these methods, the asbestos structures are identified, sized, and counted by transmission electron microscopy (TEM). Test Methods D5755 and D6480 provide results in terms of the number of asbestos structures per square centimeter of surface sampled (str/cm^2). Test Method D5756 provides results either in terms of the mass of asbestos per unit area of surface, or as the weight percent of asbestos in the dust. Micro-vacuum samples tend to more accurately reflect potential re-entrainable asbestos, while wipe samples tend to more accurately reflect all accumulated asbestos. That is, micro-vacuum sampling is more likely to remove loosely bound particles as opposed to wipe sampling that would remove particles that are more tightly bound to the surface.

FIG. 1—*Brick veneer, 1-in. air cavity, and concrete-masonry backup wall.*

This paper reports a field comparison of two measuring methods for quantifying the asbestos structure number surface loading on the vertical surface of a wall cavity of a high-rise building. The two methods are: Micro-vacuum and wipe sampling.

Background

The purpose of the project was to assess the potential World Trade Center (WTC) related chemical contamination in the building façade wall cavity of an 18-story office building in Lower Manhattan. The building façade has a brick veneer wall design that incorporates approximately a 1-in. air cavity and a concrete-masonry backup wall (Fig. 1).

The building owner alleged that the particulate plume generated by the Sept. 11, 2001 collapse of the WTC penetrated the brick veneer and deposited "WTC-particulate" in the wall cavity. The purpose of the environmental testing was to determine the presence of certain chemical "markers" or "indicators" within the façade cavity that have been identified in WTC-particulate" These chemical substances (such as asbestos, inorganic metals, polyaromatic hydrocarbons, polychlorinated biphenyls, polychlorinated dibenzodioxins and furans, man-made vitreous fibers, and particulate) are not unique to WTC-particulate. In fact, they are ubiquitous in buildings because they are contained in many building components and/or generated by other daily activities outside of the

building [9,10]. For example, the analysis of seven bulk samples of wall components (including joint caulking, brick, and mortar) obtained from the subject 18-story office building contained 2 to 25 % chrysotile and trace to 15 % tremolite asbestos.

Selection of Sampling Locations

The sampling locations were selected using two techniques: Stratified random sampling and best engineering judgment (BEJ). Random sampling ensures that bias is not introduced regarding what location is selected for sampling and allows for a statistical statement regarding the data collected. Whereas, BEJ sampling is based on intuition and informed judgment; hence, BEJ is biased in selection of sampling locations based on site-specific information. For example, the contamination may more likely be present in the wall cavity at locations where the brick veneer displays physical deterioration such as missing or damaged caulking or masonry joint mortar. A paper by Seiler et al. (1987) compares the two sampling techniques for assessment surface chemical contamination in office buildings [11].

Stratified Random Sampling

An imaginary grid of nine equal areas was constructed over the 18-story brick façade (approximately 86-ft wide by 220-ft high). The building façade was then divided vertically and horizontally into thirds yielding three strata (top, middle, and bottom) of approximately equal size. Each of the nine areas was subdivided into eight areas (or strata) of approximately equal size. Random sampling was used to identify which stratum in each of the eight areas to sample. Each stratum had an equal and independent chance of being selected as the sampling location. The locations of the nine random sampling locations are shown in Fig. 2.

Two locations in each of the three strata were selected using BEJ sampling, yielding a total of six locations. The locations were selected at areas of the brick veneer that showed physical deterioration such as missing or damaged caulking or masonry joint mortar, or any physical condition that would facilitate entry of wind-driven particulate into the wall cavity. These areas represented the areas with the highest potential for contamination in the wall cavity from wind-driven particulate.

Sampling and Analytical Methodology

Preparation of Sample Locations

An experienced mason removed the brick veneer to yield an area of approximately 18-in. by 18-in. to facilitate inspection and sampling of the concrete-masonry backup wall (Fig. 3). The mason used techniques to remove the brick masonry that minimized the generation of dust that could potentially be deposited in the wall cavity.

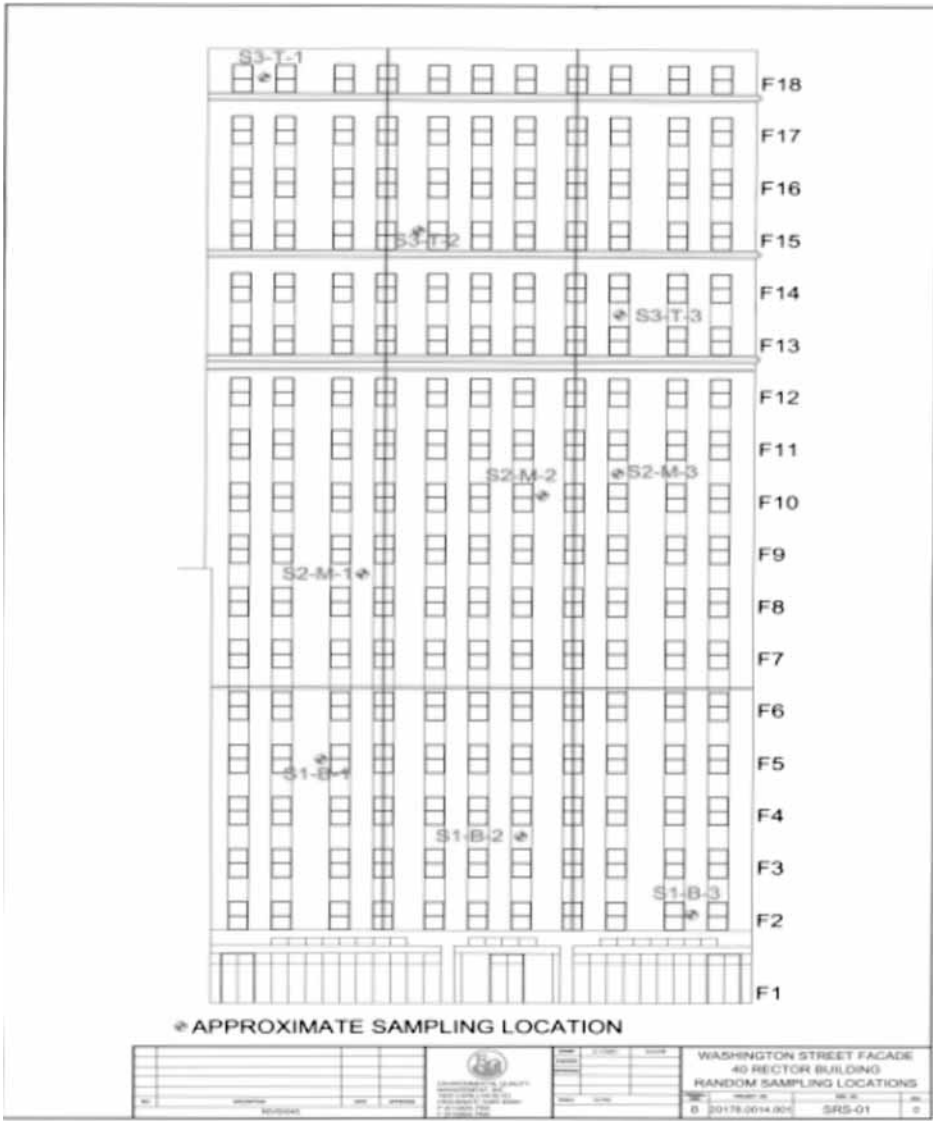

FIG. 2—*Random sampling locations on exterior façade of 18-story office building (86-ft wide by 220-ft high) BEJ sampling.*

Sample Collection and Analysis

An aluminum sample grid consisting of 16 equal 100−cm^2 areas was used as a sampling template. The template was used to ensure that equal and contiguous areas were sampled each time (Fig. 3).

FIG. 3—*Sampling template with equal 100−cm² sample areas positioned in 18-in. by 18-in. opening in brick veneer.*

Wipe Samples—The samples were collected and analyzed in accordance with ASTM Method D 6480–05, *Test Method for Wipe Sampling of Surfaces, Indirect Preparation, and Analysis for Asbestos Structure Number Loading by Transmission Electron Microscopy*. Wipe samples were collected with Ghost Wipes® pre-moistened with deionized water. A surface area of 100 cm^2 was wiped using the wiping procedure described in Section 9.2.2 of Method D6480-05.

Micro-Vacuum Samples—The samples were collected and analyzed in accordance with ASTM Method D 5755–03, *Test Method for Micro-Vacuum Sampling and Indirect Analysis of Dust by Transmission Electron Microscopy for Asbestos Structure Number Surface Loading*. The samples were collected using a three-piece air monitoring cassette containing a 37-mm diameter 0.8−μm pore size mixed-cellulose ester membrane filter and cellulose support pad. The inlet to the cassette was fitted with approximately a 25-mm-long piece of connecting tube with an edge cut at about a 45° angle. The cassette assembly was attached to a calibrated constant-flow sampling pump operating at 2 ± 0.1 l/min. The samples were collected from a known surface area of 100 cm^2. The area was sampled in two orthogonal passes for 2 min as determined by a stopwatch.

TABLE 1—*Descriptive statistics for asbestos concentrations in surface dust, structures/cm²*.

Type Sample	*N*	Mean	95 % CI	Median	Minimum	Maximum
Wipe	15	8000	5735	5500	500	35000
Micro-vacuum	15	74185	128100	5600	700	840000

Quality Control Samples—The samples were collected to assess the sampling and analytical processes and to ensure that these processes were being conducted properly. Quality control samples included field blanks and duplicate samples. Sample chain-of-custody procedures were in accordance with ASTM Method D4850–95.

Statistical Analysis

The data were analyzed to determine the mean, 95 % confidence interval, median, minimum, and maximum concentration values. The data were assumed to follow a lognormal distribution [12] and were transformed by taking the natural logarithm of each sample concentration before calculating the difference in asbestos concentration between the wipe and micro-vacuum samples. The log transformed data were compared using the parametric *t*-test. Although the distributional assumptions of normality and equal variance associated with the *t*-test analysis were reasonable, the data were also analyzed using the nonparametric alternative to the *t*-test: The Mann–Whitney Rank Sum Test. The power of each test was performed with $\alpha' = 0.05$.

Half the detection limit (i.e., L/2) was used as an estimate of the concentration of asbestos in samples reported as no structures detected (i.e., below the analytical sensitivity (AS)). This approach assumes that on average all values between the detection limit and zero could be present, and that the average value of non-detects could be as high as half the detection limit.

Results and Discussion

Comparison of Micro-Vacuum to Wipe Samples—Concentrations

The descriptive statistics for the micro-vacuum and wipe dust sample analyses are summarized in Table 1. The average asbestos concentration measured by micro-vacuum sampling ($74,185\,\mathrm{str/cm^2}$) was approximately nine times higher than that measured by wipe sampling ($8,000\,\mathrm{str/cm^2}$). The coefficient of variation is 3.0 and 1.2, respectively, which suggests a wider dispersion of the asbestos concentrations measured by micro-vacuum sampling than by wipe sampling. The respective median values ($5500\,\mathrm{str/cm^2}$ and $5600\,\mathrm{str/cm^2}$) are much more representative of the central tendency of the data and suggest that the concentrations are similar.

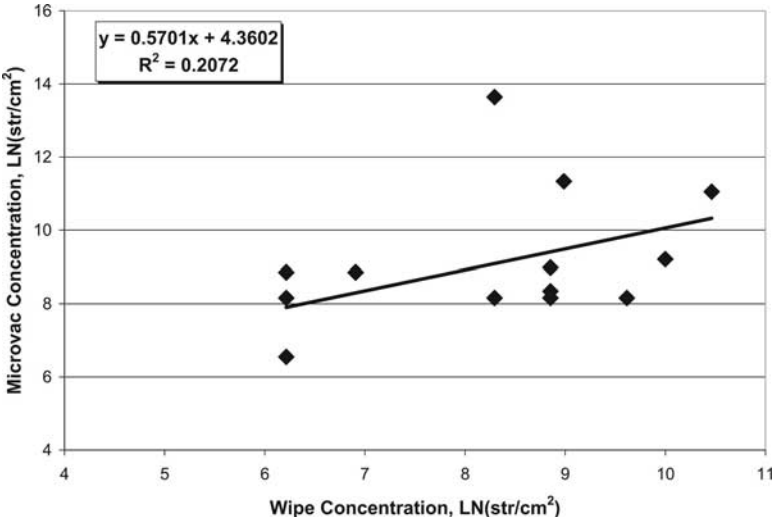

FIG. 4—*Natural logarithmic scatter plot for side-by-side micro-vacuum wipe pairs.*

To determine whether the magnitude of the difference between the average asbestos concentrations measured by the two sampling methods was significantly different, the paired-data sets were log transformed and analyzed. The log transformed data sets passed the normality ($p = 0.658$) and equal variance test ($p = 0.928$). The *t*-test showed that the difference in the mean concentrations was not significantly different ($p = 0.195$). Although the distributional assumptions of normality and equal variance were reasonable, the paired-data set was also analyzed using the Mann–Whitney Rank Sum Test. The Rank Sum Test does not require assumptions regarding the shape of the underlying distribution because it analyzes the relative ranks of the data rather than the actual measurements. The result of the Rank Sum Test is consistent with the parametric *t*-test, showing that the difference in median asbestos concentration is not significantly different ($p = 0.432$).

The lognormal distribution is the most commonly used probability density model for environmental data [12]. Hence, the results of the side-by-side sampling showing the relationship between the wipe method and micro-vacuum method are presented using a logarithmic scale (Fig. 4). The coefficient of determination (R^2) for the fit of this data to the regression line is 0.207, which does not represent an acceptable correlation. That is, a perfect correspondence between the data would have a slope of 1, an intercept of zero, and a R^2 of 1.

The results given by the surface dust measuring methods did not correlate well with each other (Fig. 4). One reason is likely the absence of homogeneity of the asbestos-containing dust distribution on the concrete-masonry wall surface in the sampled areas. Another reason is the difference in collection methods. Micro-vacuum sampling collects particles by suction, while wipe sampling collects particles by mechanical lifting. Some surfaces can trap particles in ways that affect the relative efficiency of particle collection by these two methods.

TABLE 2—*Frequency of micro-vacuum-wipe sample combination pairs with detectable and non-detectable concentrations of asbestos.*

Micro-vacuum	Wipe	% of Occurrences
Non-detect	Non-detect	0
Detect	Detect	47
Non-detect	Detect	27
Detect	Non-detect	27

That is, one would expect to wipe a smaller percentage of particles from a rough surface because particles are more likely to be deposited in surface pores and crevices [13].

The adhesion force between an asbestos particle and surface determines whether the particle is removed during sampling by either method. Hecht (1990) reports that adhesive forces are a function of particle size [14]. For the most part, adhesive strength between particles and a surface are due to Van der Waals, electrostatic, and capillarity forces, yet predominance of one or more at the same time depends on environmental conditions (such as humidity) during the sampling and physical-chemistry properties of the materials in contact [15,16]. Adhesive strength (F_{ad}) is acknowledged as a sum of various forces and is illustrated by Eq (1)

$$F_{\text{ad}} = F_{\text{vdw}} + F_e + F_c \tag{1}$$

where:

F_{vdw} = Van der Waal force,
F_e = electrostatic force, and
F_c = capillarity force.

Comparison of Micro-vacuum to Wipe Samples—% Samples $>$ or $<$ AS

Table 2 presents the percentage of the micro-vacuum-to-wipe paired samples with asbestos concentrations greater than or less than the limit of AS. The percent of like pairs ($0\% + 47\% = 47\%$) approximates the correspondence between methods. The contrast between the percentage of non-detect micro-vacuum samples that correspond to detectable wipe samples (27 %) and the number of non-detect wipe samples that correspond to detectable micro-vacuum samples (27 %) demonstrates the comparability of the two methods. In this paper, "detect" and "detectable" means counting of 1 asbestos structure in the analysis (i.e., the AS), whereas, "non-detect" means counting of no asbestos structures in the analysis.

Both methods are subject to producing false negatives, i.e., asbestos was measured in one sampling method and not in the other sampling method at a contiguous area. In this study, the false-negative rates (27 %) were the same for both methods. The false-negative rates may in part be due to the absence of homogeneity of the asbestos-containing dust distribution in the wall cavity.

TABLE 3—*Descriptive statistics for asbestos structure sizes, μm.*

	N	Mean	95 % CI	Median	Minimum	Maximum
Length						
Wipe	15	4.57	1.42	2.60	0.80	26.40
Micro-vacuum	15	2.37	0.40	1.60	0.60	15.20
Width						
Wipe	15	0.151	0.07	0.07	0.04	1.44
Micro-vacuum	15	0.063	0.02	0.04	0.04	1.03

Comparison of Micro-Vacuum to Wipe Samples—Structure Size

Table 3 summarizes the size (length and width) of the asbestos structures collected by the micro-vacuum and wipe samples. The average length of asbestos structures collected by wipe samples (4.574 μm) was approximately two times larger than that collected by the micro-vacuum samples (2.374 μm). Similarly, the average width of asbestos structures collected by wipe samples (0.151 μm) was approximately 2.4 times larger than that collected by the micro-vacuum samples. The aspect ratio (length-to-width) of the average structure sizes reported by the wipe (≈31:1) and micro-vacuum (≈40:1) samples shows that the asbestos structures were relatively long and thin. It is not known whether the size relationship would apply to particles of different shapes and surface textures. The size relationship was also maintained for the median length and width of the asbestos structures (Table 3).

To determine whether the magnitude of the difference between the median length and width of the asbestos structures measured by the wipe and micro-vacuum sampling methods was significantly different, the paired-data sets were analyzed using the nonparametric Rank Sum Test. The comparison test showed that the median structure length and width for the two sampling methods were significantly different ($p = <0.001$ length and $p = <0.001$ width). That is, the sizes of asbestos structures collected by wipe samples were significantly larger than those collected by micro-vacuum samples.

Conclusions

Two ASTM standard methods for measuring asbestos in settled dust were evaluated by a side-by-side field comparison: Micro-vacuum and wipe sampling. An important factor in this comparison was the likely non-uniform distribution of the asbestos-containing dust in the wall cavity. Despite variations in relative efficiency of particle collection by these methods, the data pairs demonstrate that micro-vacuum and wipe sampling techniques produced statistically equivalent results on the masonry concrete surface tested. The results of the two surface dust measuring methods did not correlate well with each other most likely due to the non-uniform distribution of the asbestos-containing dust in the wall cavity. The asbestos structures collected by surface wipe samples were significantly larger (length and width) than those collected by micro-vacuum sampling. Both

sampling methods are subject to production of false negatives; the false-negative rate in this study was the same. Other studies suggest that the wipe sampling technique may be more efficient in collecting asbestos structures from dust on smooth surfaces than micro-vacuum sampling. The sampling technique selected should take into consideration the textural characteristic of the dominant surface under evaluation.

References

[1] Hurlbut, C. S. and Williams, C. R., "The Mineralogy of Asbestos Dust," *J. Ind. Hyg.*, Vol. 17, 1935, pp. 289–297.

[2] Carter, R. F., "The Measurement of Asbestos Dust Levels in a Workshop Environment," *A.W.R.E. Report No.* 028/70, United Atomic Energy Authority, Aldermaton, Berkshire, U.K., 1970.

[3] Millette, J. R. and Hays, S. M., *Settled Asbestos Dust Sampling and Analysis*, Lewis Publishers, CRC Press, Inc., Boca Raton, FL, 1994.

[4] Crankshaw, O. S., Perkins, R. L., and Beard, M. E., "An Overview of Settled Dust Analytical Methods and Their Relative Effectiveness," *Advances in Environmental Measurement Methods for Asbestos, ASTM STP 1342*, M. E. Beard and H. L. Rook, Eds., ASTM International, West Conshohocken, PA, 2000.

[5] Ewing, W. M., "Further Observations of Settled Asbestos Dust in Buildings," in *Advances in Environmental Measurement Methods for Asbestos, ASTM STP 1342*, M. E. Beard and H. L. Rook, Eds., ASTM International, West Conshohocken, PA, 2000.

[6] Hatfield, R. L., Krewer, J. A., and Longo, W. E., "A Study of the Reproducibility of the Micro-Vac Technique as a Tool for the Assessment of Surface Contamination in Buildings with Asbestos-Containing Materials," *Advances in Environmental Measurement Methods for Asbestos, ASTM STP 1342*, M. E. Beard and H. L. Rook, Eds., ASTM International, West Conshohocken, PA, 2000.

[7] ASTM D7390-07, 2007, "Test Method for Evaluating Asbestos in Dust on Surfaces by Comparison Between Two Environments," *Annual Book of ASTM Standards*, Vol. 11.07, ASTM International, West Conshohocken, PA, pp. 1317–1331.

[8] Occupational Safety and Health Administration (OSHA), "Sampling for Surface Contamination," Chap. 2, *OSHA Technical Manual TED-1-0.15A*, OSHA, Washington, D.C.

[9] Kominsky, J. R., "Background Concentrations of Polychlorinated Dibenzofurans and Polychlorinated Dibenzo-p-Dioxins in Commercial Buildings," *Chemosphere*, Vol. 18(1–6), pp. 599–608, 1989.

[10] Kominsky, J. R., "PCB Exposures Following Fluorescent Light Ballast Burnout," *Appl. Ind. Hyg.*, Vol. 2(3), 1987, pp. R-23–R-26.

[11] Seiler, F. A., Davis, H. T., Kominsky, J. R., Ronan, R. J., and Kowka, C. D., "Use of Risk Assessment Methods in the Certification of Decontaminated Buildings," *Risk Anal.*, Vol. 7(4), 1987, pp. 487–495.

[12] Gilbert, R. O., *Statistical Methods for Environmental Pollution Monitoring*, Chap. 1, Van Nostrand Reinhold, New York, 1987.

[13] Royster, G. W. and Fish, B. R., "Techniques for Assessing Removable Surface Contamination," *Surface Contamination: Proceedings of a Conference Held in Gatlinburg Tennessee*, Gatlinburg, TN, June 8-12, 1964, Pergamon Press, Oxford.

[14] Hecht, L., "Particle Adhesion to Solid Surfaces," *J. Inst. Environ. Sci. Tech*, Vol. 33(2), 1990, pp. 33–37.

[15] Corn, M., "The Adhesion of Solid Particles to Solid Surfaces: A Review," *J. Air Pollut. Control Assoc.*, Vol. 11, 1961, pp. 523–528.

[16] Pakarinen, O. H., Foster, A. S., Paajanen, M., Kalinainen, T., Katainen, J., Makkonen, I., Lahtinen, J., and Nieminen, R. M., "Towards an Accurate Description of the Capillary Force in Nanoparticle-Surface Interactions," *Modell. Simul. Mater. Sci. Eng.*, Vol. 13, 2005, pp. 1175–1186.

Reprinted from JAI, Vol. 8, No. 8
doi:10.1520/JAI103509
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Use of the ASTM Inter-Laboratory Studies (ILS) Program in Developing Precision Data for ASTM D5755 – Asbestos in Dust by Microvacuum Sampling

ABSTRACT: ASTM Standard D5755 "Standard Test Method for Microvacuum Sampling and Indirect Analysis of Dust by Transmission Electron Microscopy (TEM) for Asbestos Structure Number Concentration" uses a microvacuuming sampling procedure and transmission electron microscopy analysis to (a) identify asbestos in surface dust and (b) provide an estimate of the surface loading of asbestos in the sampled dust reported as the number of asbestos structures per unit area of sampled surface. In a precision study of the method, ten laboratories analyzed chrysotile asbestos-containing World Trade Center dust that had been resuspended in a test room and collected off of non-asbestos floor tiles following the microvacuum cassette (microvac) collection procedures described in ASTM D5755. Each laboratory analyzed the same sample twice. The Inter-Laboratory Studies (ILS) Group of ASTM International performed the statistical examination of the data set and found a repeatability limit (r) of 61.80 and a reproducibility limit (R) of 239.30. The study data was also used to calculate coefficients of variation of 0.15 intra-laboratory and 0.6 inter-laboratory.

KEYWORDS: asbestos, settled dust, precision, WTC dust

Introduction

ASTM Standard D5755 "Standard Test Method for Microvacuum Sampling and Indirect Analysis of Dust by Transmission Electron Microscopy (TEM)

Manuscript received October 17, 2010; accepted for publication June 9, 2011; published online July 2011.

[1] MVA Scientific Consultants.

Cite as: Millette, J. R., "Use of the ASTM Inter-Laboratory Studies (ILS) Program in Developing Precision Data for ASTM D5755 – Asbestos in Dust by Microvacuum Sampling," *J. ASTM Intl.*, Vol. 8, No. 8. doi:10.1520/JAI103509.

for Asbestos Structure Number Concentration" [1] is a procedure to (a) identify asbestos in surface dust using TEM and (b) provide an estimate of the surface loading of asbestos in the sampled dust reported as the number of asbestos structures per unit area of sampled surface. This procedure uses a microvacuuming sampling technique employing an air filter cassette to which a piece of plastic tubing has been attached to the inlet orifice as a sampling nozzle. Particulates are vacuumed from a known area of a surface within a minimum of 2 min of sampling time. The sample is prepared by washing the particulate from the cassette and filtering a portion of the resulting suspension through a membrane filter. Asbestos identification by transmission electron microscopy (TEM) is based on morphology, selected area electron diffraction (SAED), and energy dispersive x-ray analysis (EDXA). Some information about structure size is also determined. The method has been used as an indicator of the amount of asbestos present in surface dust in buildings. Following the World Trade Center disaster of Sept 11, 2001, the method was used to assess the impact of the WTC dust in New York City residences. As part of their WTC Dust Clean-Up project, the USEPA set a risk-based benchmark for surface asbestos levels using ASTM D5755 of 5000 asbestos structures per square centimeter (str/cm^2) in accessible areas and 50 000 str/cm^2 in inaccessible areas. ASTM D5755 can be used to compare the relative asbestos fiber loadings in the surface dusts found in two areas using the ASTM D7390-07, "Standard Guide for Evaluating Asbestos in Dust on Surfaces by Comparison Between Two Environments."

Precision Study

The precision statement was determined through statistical examination of 20 results, from ten laboratories, on a single type of material. World Trade Center dust was used as the test material. An asbestos abatement type enclosure was constructed with two layers of 4-mil polyethylene plastic sheets on a wooden frame and three layers of 4-mil polyethylene plastic on the floor. The test chamber was approximately 8 feet long $\times$ 8 feet wide $\times$ 7 feet high. Sixteen [16] non-asbestos floor tiles were placed in a 4-tile-by-4-tile square. On each tile a pre-weighed 10 cm square piece of weighing paper was held in place with a metal template. Approximately 1 g of the WTC dust was resuspended in the test chamber using compressed air by blowing into a small container holding the dust. A leaf blower and four stationary fans directed upward were used to mix the dust in the air for 5 min. There was no exhaust from the chamber during the resuspension or mixing. A high efficiency particulate air filtration device was used to clean the air in the chamber before and after the activity. After waiting overnight, each weighing paper was carefully collected and a dust sample was collected next to it off the non-asbestos floor tiles following the microvacuum cassette (microvac) collection procedures described in ASTM D5755. The samples were collected from template areas of 100 cm^2 using a flow rate of 2 l per min. The sampling time was two minutes for each sample. The sampling

cassettes contained a 25 mm diameter mixed cellulose ester membrane filter with a pore size of 0.45 μm. The sampling time was two minutes for each sample.

The weighing paper samples were weighed using a 5-place A&D HR-202i analytical balance. All samples except one were found to be within 20 % of the mean particulate weight. The outlier and the microvacuum sample collected next to it were discarded. Three microvacuum samples were chosen at random from the 15 remaining samples. These were analyzed according to the D5755 method. The coefficient of variation (cv = std dev / mean) for the results of the 3 analyses was 0.17. Of the remaining 12 microvacuum cassettes, 10 samples were selected at random and sent to the 10 independent laboratories that were participating in the study. Each laboratory was asked to analyze the same sample twice using the D5755 method. The Inter-Laboratory Studies (ILS) Group of ASTM International performed the statistical examination of the data set. The ILS Group determined repeatability limits and reproducibility limits which are specified in ASTM Practice E 177. Some details of the statistical calculations provided by the ILS Group are shown in the Appendix of this article.

Results

The results of the precision study, in terms of repeatability limits and reproducibility limits, are presented in Table 1. At the time of the study, there was no accepted reference material suitable for determining the bias of the test method; therefore no statement on bias was made.

Discussion

The interpretation of the ILS results are as follows:

Repeatability limit (r) - Two test results obtained within one laboratory shall be judged not equivalent if they differ by more than the "r" value for that material; "r" is the interval representing the critical difference between two test results for the same material, obtained by the same operator using the same equipment on the same day in the same laboratory. In this study $r = 2.8 \times$ repeatability standard deviation (S_r).

TABLE 1—*Asbestos structures per square centimeter (× 1000).*

Material	Average $\bar{X}$	Repeatability Standard Deviation s_r	Reproducibility Standard Deviation s_R	Repeatability Limit r	Reproducibility Limit R
A	147.80	22.07	85.46	61.80	239.30

TABLE 2—*Summary of Intra- and Inter-Laboratory Studies of various versions of ASTM D5755.*

Year	Method	Notes	#Labs	Mean Str/cm^2	Stdev	CV	Ref.
1988	EPA Draft	EPA Carpet study. Microvacs collected from artificially contaminated carpet sprayed with asbestos suspension in water	3	2.5×10^4		1.66	[5]
1991	ASTM Draft 1991*	Measured amounts of settled dust from top of air distribution duct directly below asbestos-containing fireproofing placed directly into microvac cassettes	9	3.4×10^7	2.44×10^7	0.7	[6]
1991–1996	ASTM Drafts 1991-1994* and D5755-95	195 microvac dust samples collected from 38 different buildings with chrysotile/ vermiculite fireproofing	1Intralab 195 samples	1.6×10^7	1.5×10^7	1.0	[6]
1992	ASTM Draft 1992*	Carpet surface samples - Plenum dust Dry mixed with Arizona road dust & cellulose insulation deposited in chamber with leaf blower mixing air, collected by microvac	3	5.1×10^7	4.2×10^7	0.8	[7]
			3	4.7×10^8	7.3×10^8	1.6	
			3	3.3×10^8	5.5×10^8	1.7	
1992	ASTM Draft 1992*	Ceiling tile surface samples - Plenum dust dry mixed with Arizona road dust & cellulose insulation deposited in chamber with leaf blower mixing air, collected by microvac	3	7.9×10^7	8.1×10^7	1.0	[7]
			3	7.9×10^8	1.3×10^9	1.6	
			3	1.8×10^8	2.6×10^8	1.4	
			3	3.5×10^6	3.0×10^6	0.9	

TABLE 2—*Continued*

Year	Method	Notes	#Labs	Mean Str/cm^2	Stdev	CV	Ref.
1992	ASTM Draft 1992*	Wood surface samples - Plenum dust dry mixed with Arizona road dust & cellulose insulation deposited in chamber with leaf blower mixing air, collected by microvac	7	3.7×10^7	3.9×10^7	1.1	[7]
			5	1.1×10^8	1.5×10^8	1.4	
			6	2.9×10^7	5.2×10^7	1.8	
1993	ASTM Draft 1992*	EPA 'real world' carpet study. Microvacs collected from cafeteria in Gov bldg in Maryland	1 IntraLab 8 samples	4.0×10^4	3.1×10^4	0.8	[8]
1997	D5755-95	Asbestos dust mixed with building dust suspended in water then added to cassettes and dried	5	5.3×10^5	6.1×10^5	1.2	[9]
1998	D5755-95	Asbestos dust mixed with building dust suspended in water then added to cassettes and dried	3	1.1×10^5	1.4×10^5	1.3	[9]
1998	D5755-95	Asbestos dust mixed with building dust suspended in water then added to cassettes and dried	1 IntraLab 20 samples	5.6×10^4	4.1×10^4	0.7	[9]
1999	D5755-95	Asbestos dust mixed with building dust suspended added to cassettes	6	3.0×10^5	2.0×10^5	0.7	[9]
1999	D5755-95	Asbestos dust mixed with building dust suspended in water then added to cassettes and dried	7	8.4×10^4	7.5×10^4	0.9	[9]

*Note: 1991 and 1992 Drafts of the ASTM dust method had no 1 mm screen and no settling time specified. There were also other differences between the drafts and the final version of the Standard published in 1995.

Reproducibility limit (R) - Two test results shall be judged not equivalent if they differ by more than the "R" value for that material; "R" is the interval representing the critical difference between two test results for the same material, obtained by different operators using different equipment in different laboratories. In this study $r = 2.8 \times$ reproducibility standard deviation (S_R).

According to the ILS results report, any judgment in accordance with the above statements would have an approximate 95 % probability of being correct. Some statistical details are provided in the Appendix.

The ILS reported a repeatability standard deviation (intra-laboratory) value of 22.07×10^3 str/sq cm and the reproducibility (inter-laboratory) value of 85.46×10^3 str/cm^2 correspond to coefficients of variation (cv = std dev / mean) of 0.15 intra-laboratory and 0.6 inter-laboratory. The intra-laboratory CV agrees with the value of 0.2 reported by one laboratory for D5755 when they used it for analyzing over 2000 surface dust samples collected in conjunction with World Trade Center disaster [2]. It is also consistent with Crankshaw of the RTI group who concluded that the inter-sample variability for repeated analyses was typically less than ± 15 % after testing of laboratory prepared samples of asbestos fibers in different dust matrices [3]. The inter-laboratory CV value of 0.6 for the D5755 method is similar to what has been found for other asbestos counting methods. For instance, it is within the inter-laboratory CV range of 0.27–0.85 reported for the NIOSH 7400 phase contrast microscopy air fiber count method (A rules) [4].

A number of inter-laboratory precision studies of the TEM-microvac surface dust method (earlier versions of ASTM D5755) have been conducted since 1988. These earlier inter-laboratory precision studies all used either draft versions of the D5755 method or used samples prepared without the microvac surface collection portion of the method. Table 2 summarizes the data from several published and unpublished studies. Early round robin testing found CVs more in the 1.6–1.8 range and later testing results showed lower CV values (0.9–0.7). A review of these data is helpful in understanding how the inter-laboratory precision improves over time, presumably as laboratories become more familiar with the method and the method becomes more standardized.

Acknowledgment

This study was done with the cooperation of the following participating laboratories: AmeriSci Group, Midlothian, VA; Asbestech, Carmichael, CA; Bureau Veritas North American, Kennesaw, GA; E.M. Analytical, Dania, FL; Forensic Analytical, Hayward, CA; Hygeia Laboratories, Sierra Madre, CA, International Asbestos Testing, Mount Laurel, NJ; MVA Scientific Consultants, Duluth, GA; and Reservoirs Environmental, Denver, CO.

APPENDIX

TABLE 3—*The consistency statistic h is an indicator of how one laboratory's cell average, for a particular material, compares with the average of the other laboratories.The consistency statistic, k, is an indicator of how one laboratory's within-laboratory variability, under repeatability conditions, on a particular material, compares with all of the laboratories combined. Values of k larger than 1 indicate greater within-laboratory variability than the average for all laboratories. Since such variation among laboratories is expected, critical values of k have been calculated to aid in the decision of whether the cell standard deviation of one laboratory is sufficiently different from the rest of the laboratories as to require investigation. Critical Values of h and k at the 0.5 % Significance Level*[a]

		Critical values of k Number of replicates, n								
Critical value of h	p	2	3	4	5	6	7	8	9	10
1.15	3	1.72	1.67	1.61	1.56	1.52	1.49	1.47	1.44	1.42
1.49	4	1.95	1.82	1.73	1.66	1.60	1.56	1.53	1.50	1.47
1.74	5	2.11	1.92	1.79	1.71	1.65	1.60	1.56	1.53	1.50
1.92	6	2.22	1.98	1.84	1.75	1.68	1.63	1.59	1.55	1.52
2.05	7	2.30	2.03	1.87	1.77	1.70	1.65	1.60	1.57	1.54
2.15	8	2.36	2.06	1.90	1.79	1.72	1.66	1.62	1.58	1.55
2.23	9	2.41	2.09	1.92	1.81	1.73	1.67	1.62	1.59	1.56
2.29	10	2.45	2.11	1.93	1.82	1.74	1.68	1.63	1.59	1.56
2.34	11	2.49	2.13	1.94	1.83	1.75	1.69	1.64	1.60	1.57
2.38	12	2.51	2.14	1.96	1.84	1.76	1.69	1.64	1.60	1.57
2.41	13	2.54	2.15	1.96	1.84	1.76	1.70	1.65	1.61	1.58
2.44	14	2.56	2.16	1.97	1.85	1.77	1.70	1.65	1.61	1.58
2.47	15	2.57	2.17	1.98	1.86	1.77	1.71	1.66	1.62	1.58
2.49	16	2.59	2.18	1.98	1.86	1.77	1.71	1.66	1.62	1.58
2.51	17	2.60	2.19	1.99	1.86	1.78	1.71	1.66	1.62	1.59
2.53	18	2.61	2.20	1.99	1.87	1.78	1.72	1.66	1.62	1.59
2.54	19	2.62	2.20	2.00	1.87	1.78	1.72	1.67	1.62	1.59
2.56	20	2.63	2.21	2.00	1.87	1.79	1.72	1.67	1.63	1.59
2.57	21	2.64	2.21	2.00	1.88	1.79	1.72	1.67	1.63	1.59
2.58	22	2.65	2.21	2.01	1.88	1.79	1.72	1.67	1.63	1.59
2.59	23	2.66	2.22	2.01	1.88	1.79	1.72	1.67	1.63	1.59
2.60	24	2.66	2.22	2.01	1.88	1.79	1.73	1.67	1.63	1.60
2.61	25	2.67	2.23	2.01	1.88	1.79	1.73	1.67	1.63	1.60
2.62	26	2.67	2.23	2.02	1.89	1.80	1.73	1.68	1.63	1.60
2.62	27	2.68	2.23	2.02	1.89	1.80	1.73	1.68	1.63	1.60
2.63	28	2.68	2.23	2.02	1.89	1.80	1.73	1.68	1.63	1.60
2.64	29	2.69	2.24	2.02	1.89	1.80	1.73	1.68	1.64	1.60
2.64	30	2.69	2.24	2.02	1.89	1.80	1.73	1.68	1.64	1.60

[a]The above critical values for the h and k consistency statistics were calculated from Student's t and the F-ratio using the following relationships: $h = (p-1)\, t\sqrt{p(t2+p-2)t}$ with $p-2$ degrees of freedom, and $k = \sqrt{p/[1+(p-1)/F]}\, F$ with $n-1$ and $(p-1)(n-1)$ degrees of freedom. where p = number of laboratories.

References

[1] ASTM D5755-10, 2010, Standard Test Method for Microvacuum Sampling and Indirect Analysis of Dust by Transmission Electron Microscopy for Asbestos Structure Number Concentration, *Annual Book of ASTM Standards*, ASTM International, West Conshohocken, PA.

[2] Ehrenfeld, F., *International Asbestos Testing*, Mount Laurel, NJ, personal communication, 2010.

[3] Crankshaw, O., Perkins, R. L., and Beard, M. E., "An Overview of Settled Dust Analytical Methods and Their Relative Effectiveness," *Advances in Environmental Measurement Methods for Asbestos, ASTM STP 1342*, M. E. Beard and H.L. Rook, Eds., American Society for Testing and Materials, Philadelphia, 2000.

[4] NIOSH Method 7400, *Asbestos and Other Fibers by PCM, in NIOSH Manual of Analytical Methods (NMAM)*, 4th ed., 1994.

[5] Millette, J. R., Clark, P. J., Brackett, K. A., and Wheeles, R. K., "Methods for the Analysis of Carpet Samples for Asbestos," *Environ. Choices Tech. Supp.*, Vol. 1, No. 2, 1993, pp. 21–24.

[6] Hatfield, R. L., Krewer, J. A., and Longo, W. E., "A Study of the Reproducibility of the Micro-Vac Technique As A Tool for the Assessment of Surface Contamination in Buildings with Asbestos-Containing Materials," *Advances in Environmental Measurement Methods for Asbestos, ASTM STP 1342*, M. E. Beard and H. L. Rook, Eds., American Society for Testing Materials, Philadelphia, 2000.

[7] Lee, R. J., VanOrden, D. R., and Dunmyre, G. R., "Interlaboratory Evaluation of the Breakup of Asbestos-Containing Dust Particles by Ultrasonic Agitation," *Environ. Sci. Technol.*, Vol. 30, No. 10, 1996, pp. 3010–3015.

[8] Kominsky, J. R., Freyberg, R. W., Chesson, J., Cain, W. C., Powers, T. J., and Wilmoth, R. C., "Evaluation of Two Cleaning Methods for the Removal of Asbestos Fibers from Carpet," *Am. Ind. Hyg. Assoc. J.*, Vol. 51, No. 9, 1990, pp. 500–504.

[9] Millette, J. R., ASTM D22.07, Subcommittee, Results of Preliminary Round Robin Testing of D5755, 1997–1999.

PHARMACEUTICALS

Reprinted from JAI, Vol. 8, No. 8
doi:10.1520/JAI103480
Available online at www.astm.org/JAI

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Developing Acceptable Surface Limits for Occupational Exposure to Pharmaceutical Substances

ABSTRACT: Acceptable surface limits (ASLs) are developed in order to establish a quantitative measure for the potential risk from exposure by dermal contact. In the pharmaceuticals industry, ASLs are used for protection against active pharmaceutical ingredients that are known to cause pharmacological or toxicological effects. An ASL can be used, together with appropriate analytical methods and industrial hygiene monitoring, to assess workplaces for potential dermal exposure and to protect the health and safety of individuals who might come in direct contact with contaminated surfaces in the workplace. ASLs are also used to evaluate the adequacy of housekeeping measures and the effectiveness of engineering containment approaches, or to determine whether a chemical is present on surfaces where it is not intended to be (e.g., in lunch rooms or offices, or on the outside surfaces of packaging materials). However, they should not be confused with cleaning limits for the surfaces of manufacturing devices that might come into contact with the drug product, which are set to minimize cross contamination between drug products and to protect end-users (e.g., patients taking drug products) as opposed to workers. A number of parameters must be evaluated in order to accurately develop appropriate and scientifically supportable limits. These include the dose or concentration that will cause the potential effect, the degree of chemical transfer from contaminated surfaces to the

Manuscript received October 12, 2010; accepted for publication June 26, 2011; published online August 2011.

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Cite as: Kimmel, T. A., Sussman, R. G., Ku, R. H. and Ader, A. W., "Developing Acceptable Surface Limits for Occupational Exposure to Pharmaceutical Substances," *J. ASTM Intl.*, Vol. 8, No. 8. doi:10.1520/JAI103480.

skin, and the rate or amount of percutaneous absorption. In practice, this information is usually limited or unavailable. Additionally, there has been very little regulatory guidance on the setting of ASLs. Consequently, in order to calculate an ASL, various assumptions must be made by health and safety professionals regarding how dermal exposures might occur. As quantitative data become available, the ASL can be adjusted accordingly. An overview of the setting of health-based and performance-based ASLs for pharmaceutical substances from animal and human toxicological data is provided.

KEYWORDS: surface limit, occupational, pharmaceuticals

Introduction

In the workplace, systemic exposure to chemical substances occurs primarily via the inhalation route. However, another important route of occupational exposure is through dermal contact. During manufacturing operations, surfaces can become contaminated for several reasons, including inadequate containment and/or cleanup processes following spills, ineffective enclosures and engineering controls, poor employee training on industrial hygiene practices, inadequate and/or improper housekeeping practices, and specific physicochemical processes of the chemical(s) being handled. During the manufacture of active pharmaceutical ingredients (APIs), there are additional challenges related to a known or predicted pharmacological response following a systemic exposure, which, regardless of its nature, would be considered deleterious to a healthy worker population. Moreover, certain APIs that are developed for transdermal administration have inherent dermal absorptive properties or are mixed with permeation-enhancing chemicals in topical formulations in order to facilitate systemic exposure. Consideration must also be given to potency, which is becoming increasingly high as new drugs are being investigated/marketed that are pharmacologically active at low therapeutic doses. As a result, even the smallest exposures to these novel APIs via any route can potentially present a serious health risk.

As mentioned previously, APIs are intended to be pharmacologically active, but there are also other characteristics that distinguish APIs from commodity chemicals. APIs, although biologically reactive, tend to be chemically stable (note: the same might not be true of their synthetic intermediates). In addition, most APIs are solids and as such are readily deposited on surfaces, whereas commodity chemicals might be solid, liquid, or gas. APIs tend to have more robust toxicokinetic/toxicodynamic profiles than commodity chemicals. These data are collected as part of the drug research and development process and include critical information such as the systemic bioavailability and bioaccumulation potential. Frequently, there is a wealth of human data available for an API, whereas for other chemicals the human data are often limited to accidental (over)exposures with resultant toxicities. Overall, the data set that is routinely assembled for an API is better suited for performing human health risk assessments.

Factors Influencing Dermal Absorption

The process of chemical migration from the surface of the skin to the systemic circulation is a complex one [1,2]. There are many factors that contribute to the dermal absorption potential of a chemical, including the following:

- *ability to penetrate the skin*, determined by such factors as physical adherence to skin, the condition and thickness of the contacted skin, the number of sweat glands and hair follicles at the site of contact (even though these make very small contributions to the exposure), the ambient temperature in the work area, occlusion of the exposed area by clothing or other personal protective equipment (which might prolong the contact between the chemical and the skin), and inherent physicochemical properties such as the molecular size (smaller molecules are more likely to penetrate the skin) and lipophilicity (a log P_{ow} between $+1$ and $+2$ is the most favorable for dermal absorption);
- *amount of chemical that contacts the skin*, referring to the chemical concentration on the surface;
- *amount of skin that contacts the chemical*, referring to the surface area of the skin that contacts the chemical;
- *frequency and duration of the contact event*;
- *concomitant exposure to multiple chemicals*—e.g., in drug formulations (which might include permeation-enhancers); and
- *interindividual variability*.

Each of these parameters must be considered when assessing the potential for skin absorption.

Setting Dermal Exposure Limits for APIs in the Workplace

Occupational dermal exposure limits, hereinafter referred to as acceptable surface limits (ASLs), are risk assessment tools used for worker safety. Similar to occupational exposure limits, which set acceptable airborne concentrations for inhalation exposure to chemicals, the ASL is considered as a surface concentration that is "reasonably" safe to individuals following contact with unprotected skin. It is used by health and safety professionals to help determine whether it is safe for workers to re-enter a previously contaminated work area without personal protective equipment, as well as for risk communication purposes. It must be emphasized that an ASL is not equivalent to an equipment cleaning limit: ASLs are set for worker safety, whereas equipment cleaning limits (used to minimize product cross-contamination between batches of two different drug products sharing the same equipment) are intended to protect patients as opposed to workers.

An ASL cannot be properly used without the development of validated analytical methods for monitoring specific surfaces, as different surfaces might not be equally suitable for recovery of the material. A validated method requires putting a known amount on the surface, using a collection technique (sometimes called a wipe or surface sampling method) for taking the sample, and quantitatively measuring the recovered amount from that surface.

With an ASL and a validated surface sampling and analytical method, an industrial hygienist or other trained occupational health professional can conduct a workplace exposure assessment. This involves developing a plan to determine sites at which dermal contact is most likely to occur, based on the nature of the task and work patterns, and selecting appropriate areas and surfaces to be sampled.

Methodology

The pivotal step when performing a proper risk assessment for setting an ASL is to know, or be able to predict, the systemic bioavailability of a chemical via the dermal route. Limited regulatory guidance on dermal exposures is readily found in the public domain; the Environmental Protection Agency has published guidelines for estimating dermal exposures to pesticides and contaminated soil/water [2–5], and the European Union has issued a guidance document for predicting dermal exposure (with proposed revisions submitted by the UK Health & Safety Executive) that focuses on agrochemical scenarios [6,7]. Exposures in workplace settings are not expressly covered by these documents. Moreover, information on dermal absorption found in the public domain is usually compound-specific and cannot be used to extrapolate to other chemicals. Although there are new methods published for the theoretical or practical determination of individual parameters contributing to absorption (such as the flux rate and permeation coefficient), there still remains a shortage of overall guidance on understanding the entirety of the dermal absorption process. This presents a tremendous challenge in performing risk assessments.

Another difficulty in establishing quantitative dermal exposure limits for an occupational setting is that wherever exposure via the dermal route is possible, exposure via ingestion (e.g, following incidental hand-to-mouth contact) and/or inhalation is also possible. Consequently, if workers experience an adverse effect, the relative contribution of dermal exposure is unknown. For simplicity, it is assumed in this paper that dermal exposure is the only route of exposure.

Despite the lack of guidance, it is necessary for industry to develop approaches to ASL setting in order to properly protect workers. Within the pharmaceuticals industry, a common practice is to develop ASLs by performing a health-based risk assessment using readily available data and calculate an "occupational acceptable daily exposure" (ADE_{occ}) as follows:

$$ADE_{occ}(\mu g/day) = \frac{(NOAEL)(BW)}{(UF_{1,23})(\alpha)} \quad (1)$$

where:

NOAEL = no-observed-adverse-effect level for the critical endpoint of concern (if a NOAEL is not identified, a lowest-observed-adverse-effect level [LOAEL] may be selected instead),

BW = body weight (50 kg for an adult worker),

$UF_{1,2,3}$ = composite uncertainty factors for considerations such as inter-individual variability, inter-species variability, pharmacokinetic variability,

extrapolation from a LOAEL to a NOAEL, severity of adverse effects, sensitive subpopulations, and robustness of the data set, and

α = adjustment factor for bioavailability via the route of administration by which the critical effect was observed.

For APIs, ADE_{occ} may also be derived using a low therapeutic dose, expressed in mg/day or μg/day. Because a pharmacological effect in healthy workers is considered to be adverse, this dose is considered to be a LOAEL.

The ASL is then derived from the ADE_{occ} as follows:

$$ASL(\mu g/cm^2) = ADE_{occ}\frac{(\mu g/day)}{(SA)(\alpha_d)} \quad (2)$$

where:

SA = surface area of the skin that comes into contact with the API each day, and

α_d = adjustment factor for bioavailability via the dermal route of exposure.

For this approach, it is assumed that the average surface area of each palm is 100 cm^2, the total area of contact is equivalent on average to two palms (200 cm^2), and, in the absence of data to suggest otherwise, dermal transfer (adherence and absorption) is complete (100%). These assumptions reflect the highly conservative and protective nature of this approach, which is needed given that the process of dermal absorption remains poorly characterized. The area that is typically sampled by the industrial hygienist when monitoring potential surface contamination is 100 cm^2. However, when the surface does not lend itself to using a 10 cm $\times$ 10 cm template (e.g., sampling a door handle or product vial), the surface area sampled is estimated. ASLs are expressed as mass units per square centimeter in order to account for this variability in sampled surfaces. The sampling results are compared to the ASL for the purpose of allowing workers to use less, or remove, dermal protection.

An alternative approach to the above-described health-based approach in the setting of ASLs for pharmaceuticals is to "align" the ASL with an occupational health categorization ("banding") system used for both hazard characterization and exposure control. Each category is determined by toxicological and potency criteria for the intrinsic hazard of an API; the category to which an API is assigned increases relative to the degree of hazard. Categories can also be associated with a range of (inhalation) occupational exposure limit values, within which a value is likely to be calculated for the API and similarly may be linked to ASLs; higher categories correspond to lower exposure limits. As an example, a 4-band categorization system is described as follows:

- category 1 is assigned to APIs of low toxicity and/or potency.
- category 2 is assigned to APIs of low-to-moderate toxicity and/or potency.
- category 3 is assigned to APIs of high toxicity and/or potency.
- category 4 is assigned to APIs of very high toxicity and/or potency.

Although "potency" is a relative term ("potent" APIs are those that exert their effect at "low" dosages), a general rule used in the industry is that an API is considered to be potent if it has a pharmacologically effective or therapeutic dosage of $\leq$10 mg/day. Exposure controls and handling practices are

established based on the categorization of an API and the task(s) being performed. (For a more detailed, general discussion of the principles of a banding and handling practice system described above, the reader is referred to Ader et al. [8], Farris et al. [9], and/or Naumann et al. [10].)

According to this approach, which may be described as "performance-based" rather than health-based, ASLs may then be assigned as follows:

- For category 1 and category 2 APIs, surfaces should be sufficiently clean such that there is no visible dustiness (corresponding to an ASL of 1 to 5 $\mu g/cm^2$ [11]).
- For category 3 APIs, the ASL is an order of magnitude lower than that for category 1 or 2 APIs (0.1 to 0.5 $\mu g/cm^2$).
- For category 4 APIs, the ASL is an order of magnitude lower than that for band 3 APIs (0.01 to 0.05 $\mu g/cm^2$).

In addition, up to tenfold adjustments to the ASL for category 2 through category 4 APIs may be made based on special considerations for the API in question and/or the application of professional judgment by a properly trained occupational toxicologist. Category 2 ASLs may be adjusted downward only, whereas category 3 or 4 ASLs may be adjusted upward or downward. Some factors that contribute to the adjustment of ASLs are outlined in Table 1.

As an example, if an API is considered to fall into category 3 with an ASL of 0.1 to 0.5 $\mu g/cm^2$ but there are ancillary skin absorption data that generally support the likelihood for significant systemic absorption, then the ASL might be reduced to 0.05 $\mu g/cm^2$.

It should be noted that the purpose of the performance-based ASL (PB-ASL) system is to assign ASLs for compounds with limited data or with only a band. It is not intended to be used as a tool for determining the band based on an existing ASL. For instance, if an API is categorized in band 4, it would be assigned a PB-ASL of between 0.01 and 0.05 $\mu g/cm^2$, depending on the other

TABLE 1—*Some parameters that might be considered when adjusting a performance-based ASL.*

Criterion	May increase PB-ASL if…	May decrease PB-ASL if…
Ancillary skin absorption data[a]	Suggest that absorption is poor	Suggest that absorption is good
Molecular weight (MW)	>1000 Dalton[b]	<500 Dalton[c]
Octanol:water partition coefficient (log P_{ow})	Less than +1 or greater than +2[b]	Between +1 and +2[c]
Nature of observed toxic or adverse effects	Mild/moderate in severity and/or reversible	Severe and/or irreversible
Banding characteristics	Closer to Band 2 than 4	Closer to Band 4 than 2

[a] May include animal data, human cadaver data, experience with related compounds, predictive modeling, workplace case reports, etc.
[b] Dermal absorption is less favorable.
[c] Dermal absorption is more favorable.

variables listed in the table. It should also be noted that other factors such as chemical-to-skin contact, the frequency/duration of the contact event, and interindividual variability are not considered in either the health-based or the performance-based approach because conservative assumptions are made during the derivation (e.g., skin permeability is complete in the absence of quantitative data) and/or appropriate data are unavailable for making scientifically defensible adjustments (e.g., to account for interindividual variability). Mathematical models are available for the prediction of dermal absorption; however, they are highly complex and difficult to incorporate as part of the ASL-setting process [2,3,12–14].

Both the health-based and the performance-based ASL approaches have strengths and limitations. The health-based approach is based on a more comprehensive evaluation of the data; however, it frequently carries a high degree of uncertainty, necessitating the use of very conservative safety factors. The performance-based approach bypasses the above-mentioned conservatism by using "fixed" values based on a hazard category; however, the ASLs are assigned as ranges of values, and professional toxicological judgment is required in order to choose an appropriate limit. To the knowledge of the authors, at present there is no known "complete" approach to setting ASLs within the pharmaceutical industry or within other industries.

Summary

The potential for systemic toxicity following dermal exposure is an important consideration when assessing the hazards of a workplace setting. Active pharmaceutical ingredients have characteristics that, compared to other chemicals, can make it easier for the occupational health professional to perform a risk assessment for dermal exposure. However, the complexity of the dermal absorption process, along with limited regulatory guidance, presents challenges. At the present time, the setting of acceptable surface levels for active pharmaceutical ingredients relies on approaches that are highly conservative and/or only partially health-based. As our overall knowledge increases and reliable/validated tools for predicting or quantifying skin absorption are developed, it is anticipated that these approaches will be modified so as to increase their accuracy, appropriateness, and scientific supportability.

References

[1] Degim, I. T., "New Tools and Approaches for Predicting Skin Permeability," *Drug Discovery Today*, Vol. 11(11/12), 2006, pp. 517–523.

[2] Exposure Assessment Group, Office of Environmental Assessment, U.S. Environmental Protection Agency, 1992, "Dermal Exposure Assessment: Principles and Applications," Interim Report EPA/600/8-91/011B, oaspub.epa.gov/eims/eimscomm.getfile?p_download_id=438674 (Last accessed 1 Oct 2010).

[3] National Center for Environmental Assessment, Office of Research and Development, U.S. Environmental Protection Agency, 2007, "Dermal Exposure Assessment:

A Summary of EPA Approaches," EPA/600/R-07/040F, oaspub.epa.gov/eims/eimscomm.getfile?p_download_id=469581 (Last accessed 1 Oct 2010).

[4] U.S. Environmental Protection Agency, 1997, "Exposure Factors Handbook (Final Report) 1997," EPA/600/P-95/002F a-c, http://cfpub.epa.gov/ncea/cfm/recordisplay.cfm?deid=12464#Download (Last accessed 1 Oct 2010).

[5] American Industrial Health Council, "Dermal," *AIHC Exposure Factors Sourcebook*, The American Industrial Health Council, Washington, D.C., 1994.

[6] Health & Consumer Protection Directorate-General, European Commission, 2004, "Guidance Document on Dermal Absorption," http://ec.europa.eu/food/plant/protection/evaluation/guidance/wrkdoc20_rev_en.pdf (Last accessed 1 Oct 2010).

[7] European Food Safety Authority, 2010, "EFSA External Report: Proposal for a Revision of the Guidance Document on Dermal Absorption (Question No. EFSA-Q-2009-00554)," http://www.efsa.europa.eu/fr/scdocs/scdoc/52e.htm (Last accessed 1 Oct 2010).

[8] Ader, A. W., Farris, J. P. and Ku, R. H. "Occupational Health Categorization Handling Practice Systems—Roots, Application and Future," *Chemical Health and Safety*, Vol. 12(4), 2005, pp. 20–26.

[9] Farris, J. P., Ader, A. W., and Ku, R. H., "History, Implementation and Evolution of the Pharmaceutical Hazard Categorization and Control System," *Chemistry Today*, Vol. 24(2), 2006, pp. 5–10.

[10] Naumann, B. D., Sargent, E. V., Starkman, B. S., Fraser, W. J., Becker, G. T., and Kirk, G. D., "Performance-Based Exposure Control Limits for Pharmaceutical Active Ingredients," *Am. Ind. Hyg. Assoc. J.*, Vol. 1, 1996, pp. 33–42.

[11] Fourman, G. L. and Mullen, M. V., "Determining Cleaning Validation Acceptance Limits for Pharmaceutical Manufacturing Operations," *Pharmaceutical Technology*, Vol. 17(4), 1993, pp. 54–60.

[12] Paull, J. M., 1997, "A Proposed Risk-Based Model for the Evaluation of Surface Contamination, and the Assessment of Potential Dermal Exposure," Ph.D. thesis, School of Hygiene and Public Health, Johns Hopkins University, Baltimore.

[13] Fiserova-Bergerova, V., and Pierce, T., "Dermal Absorption," *Topics in Biological Monitoring, ACGIH BEI Committee*, ACGIH, Cincinnati, 1995, pp. 33–44.

[14] Bronaugh, R. L. and Barton, C. N., "Prediction of Human Percutaneous Absorption with Physicochemical Data," *Health Risk Assessment, Dermal and Inhalation Exposure and Absorption of Toxicants*, R. G. M. Wang, J. B. Knaak, and H. I. Maibach, Eds., CRC, Boca Raton, FL, 1992, pp. 117–134.

Reprinted from JAI, Vol. 8, No. 6
doi:10.1520/JAI103544
Available online at www.astm.org/JAI

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Development of a Method for Screening Spill and Leakage of Antibiotics on Surfaces Based on Wipe Sampling and HPLC-MS/MS Analysis

ABSTRACT: A screening method for determination of spill and leakage of 12 different antibiotic substances has been developed. The method is based on wipe sampling where the sampling procedure has been simplified for screening purposes. After sample processing, the antibiotic substances are determined by liquid chromatography coupled to tandem mass spectrometry (HPLC-MS/MS). Twelve antibiotic substances can be determined in the screening method: Cefadroxil, Cefalexin, Ciprofloxacin, Demeclocyklin HCl, Diaveridin, Doxycyklin, Enrofloxacin, Flukonazol, Metronidazol, Norfloxacin, Ofloxacin, and Trimetoprim. These substances are active components in antibiotic drugs frequently used in Sweden. For screening investigations using collection of wipe samples, good or acceptable performance was obtained for ten substances on three or more surface materials. Although not fully acceptable, useful performance for screening purposes was also obtained on the other surface materials and for the other substances, except Demeclocyklin HCl, on all surface materials. By employing a classification procedure, where the samples are divided into groups according to increasing contamination of the sample surfaces, screening samples and the contamination level can simply be compared. This classification procedure will also help to circumvent any deficiency in recovery performance for some substances and surface materials.

KEYWORDS: antibiotics, HPLC-MS/MS, leakage, occupational exposure, screening, spill, surface contamination, wipe sampling

Manuscript received November 2, 2010; accepted for publication April 29, 2011; published online July 2011.

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Cite as: Nygren, O. and Lindahl, R., "Development of a Method for Screening Spill and Leakage of Antibiotics on Surfaces Based on Wipe Sampling and HPLC-MS/MS Analysis," *J. ASTM Intl.*, Vol. 8, No. 6. doi:10.1520/JAI103544.

Introduction

Undesired spill and leakage can occur during handling of drugs in hospitals, and the medical staff may be occupationally exposed to these drugs. During the past 15 years, several studies have been published with focus on monitoring spills and leakage as well as occupational exposure to antineoplastic drugs. Dr. Tom Connor at the National Institute for Occupational Safety and Health (NIOSH) in the US has compiled a web-based comprehensive database of such literature [1]. Since the handling of antineoplastic drugs may cause exposure, work with these drugs is consequently strictly regulated in most countries [2–5]. Although each country has their own provisions, they all have a common core of regulations, i.e., all handling should be carried out in safety boxes, isolators or using closed systems, protective clothes shall be worn, special cleaning routines shall be implemented, waste shall be handled as hazardous waste, the compounding systems shall be leak-tested on a regular basis, the staff shall receive adequate education for their work, etc.

Antibiotics belong to another heterogenic group of drugs that is frequently used. In Sweden, more than 140 times more antibiotics are administered to hospital patients, compared to antineoplastic drugs [6]. Moreover, there are only limited regulations for safe handling of antibiotics in medical care, compared to antineoplastic drugs. It is therefore realistic to assume that the spill and leakage of antibiotics is the same or larger than with antineoplastic drugs. There are, however, almost no studies on spill and leakage of antibiotics in medical care.

To carry out relevant studies, there is a need for adequate screening methods to monitor spill and leakage of antibiotics at low levels. There are no methods available for such screening studies. Reversed phase liquid chromatography coupled with mass spectrometry (HPLC-MS) has, however, frequently been used for determination of many antibiotic substances in sewers and waste waters [7,8]. Similar methods have also been developed for screening spill and leakage of antineoplastic drugs. These latter screening methods are frequently based on wipe sampling followed by gas chromatography-mass spectrometry (GC-MS) [9], liquid chromatography-tandem mass spectrometry (HPLC-MS/MS) [10–12], or voltammetry [13,14] (see Ref 1 for more references).

The aim of this study has been to develop and validate a screening method for antibiotic substances, taking its basics from a previously developed method for determination of cyclophosphamide (CP) and ifosfamide (iF), based on wipe sampling followed by an HPLC-MS/MS analysis [11,15,16] of the samples. The method will be validated for screening a large number of substances used as active components in frequently used antibiotics in wipe samples collected from various frequently occurring surface materials.

Material and Methods

Material and Chemicals

All chemicals were of analytical grade or higher quality and the water was purified in a Milli-Q water purifier (Millipore Corp., Billerica, MA, US). Table 1

TABLE 1—*The antibiotic substances (in alphabetic order) considered for the method.*

Antibiotic substance	I = Included; R = rejected	Comment
Amoxicillin	R	CA
Benzylpenicillin	R	CA
Cefadroxil	I	
Cefalexin	I	
Cefotaxim	R	TLS
Ceftadizim	R	CA
Cefuroxim	R	CA
Ciprofloxacin	I	
Demeclocykline HCl	I	
Diaveridin	I	
Doxycyklin	I	
Enrofloxacin	I	
Flukonazol,	I	
Gentamicin	R	CA
Imipenem	R	TLS
Klindamycin	R	CA
Meropenem	R	TLS
Metronidazol	I	
Norfloxacin	I	
Ofloxacin	I	
Penicillins, e.g., Kloaxcillin	R	CA
Piperacillin/Tazobaktam	R	CA
Pivmecillinam	R	CA
Tobramycin	R	CA
Trimetoprim	I	
Vancomycine	R	CA

Note: The trivial names and spelling are given according to FASS [17]. CA, could not be analysed (poor chromatography or unsatisfactory MS resolution); TLS, too low sensitivity.

shows the antibiotic substances that were considered for the method, with trivial names according to FASS [17]. A total of 26 different substances were investigated. Twelve of the investigated substances were found to be possible to analyze simultaneously. These substances are presented in Fig 1, with trivial and chemical names and structural formulas according to FASS [17]. As internal standards, the following isotope-labeled substances were used: Enrofloxacin-D5 (Sigma-Aldrich, Seelze, Germany), 5.5 μg/mL in methanol, and Flukonazol-D4 (CDN Isotopes, Pointe-Claire, Canada), 4.8 μg/mL in methanol.

A wipe sampling tissue (Apoliva, Apoteket AB, Stockholm, Sweden), previously used and tested for antineoplastic materials [15,16], was employed for collecting wipe samples. The Apoliva tissue is a commercial wet tissue single

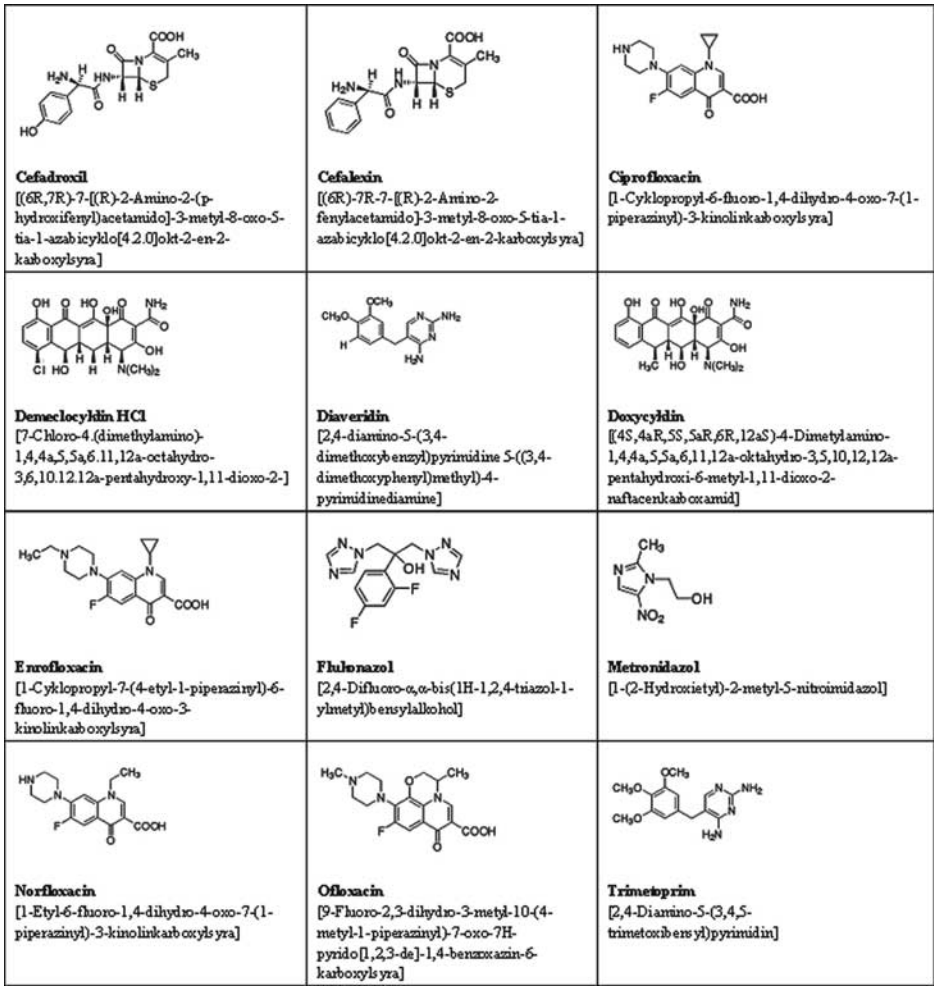

FIG. 1—*Substances included in the screening method. Structural formulas, trivial and chemical names are given according to FASS [17].*

packed in envelopes. It is a non-woven cellulose fiber tissue (17 cm × 22 cm) wetted with 3.2 g of a 15% ethanol in water solution with sorbic acid as preservative. The samples were stored in 15 mL screw-capped plastic test tubes (Sarstedt, Nümbrecht, Germany). Powder-free disposal gloves were used when taking samples.

In the sampling efficiency tests, some frequently occurring surface materials were tested. The following four materials were included in the test: a square of standard window glass (3 mm × 600 mm × 600 mm), a square (600 mm × 600 mm) from a PVC homogene plastic floor carpet roll (Armstrong, Holmsund, Sweden), a square (1.25 mm × 600 mm × 600 mm) of a stainless steel sheet used

for sinks (18/8 steel SS2332), a square (600 mm × 600 mm) from a standard laminate bench top with a melamine surface.

Instrumentation

A Perkin Elmer (Norwalk, CT, USA) chromatographic system (HPLC) consisting of two micro-pumps and an auto-sampler (Perkin Elmer series 200) was used for the analysis. The HPLC was equipped with an YMC Hydrosphere C18 column, 5 μm, 150 mm × 4.6 mm id (YMC Inc., Wilmington, NC, US). Acetonitrile in water, with 0.1% of formic acid, was used as eluent, starting at 15% acetonitrile for 2 min followed by a gradient up to 70% acetonitrile after 12 min. The HPLC system was then reset to 15% acetonitrile and equilibrated for 8 min between each run. The eluent flow rate was 0.3 mL/min. The HPLC system was coupled to a tandem mass spectrometer (MS/MS) with a triple quadrupole (API 2000 PE Biosystem, Foster City, CA, USA) equipped with an electrospray ion source (TurboIonSpray). The ion spray voltage (IS) was set to 5.5 kV (positive mode) and the drying gas (TEM) was at 350°C. Other fixed parameters were Cur (curtain gas) 20, CAD (collision gas) 5, GS1 (ion source gas 1) 20, and GS2 (ion source gas 2) 50. Table 2 shows the component specific MS/MS settings for the 12 antibiotic substances.

Wipe Sampling Procedure

Wipe samples were collected on defined areas on the selected surfaces using a wet tissue and disposal gloves. The gloves were changed between each sample to avoid cross-contamination between samples. A homemade plastic frame, encompassing 10 cm × 10 cm (= 100 cm^2), was used to sample a reproducible

TABLE 2—MS/MS *settings.*

	Q1 m/z	Q3 m/z	DP	FP	EP	CEP	CXP	CE
Cefadroxil	364	208	6	370	5	24	4	17
Cefalexin	348	158	1	360	5.5	20	2	21
Ciprofloxacin	332	288	101	330	4.5	22	8	19
Demeclocyklin HCl	465	448	21	370	8.5	18	4	27
Diaveridin	261	245	16	370	8	26	10	25
Doxycyklin	445	428	31	370	10	20	4	33
Enrofloxacin	360	316	16	370	5.5	28	10	27
Flukonazol	307	220	26	320	9	22	6	27
Metronidazol	172	154	26	370	6	26	4	23
Norfloxacin	320	276	21	370	7	18	4	27
Ofloxacin	362	318	26	370	8.5	22	4	25
Trimetoprim	291	230	36	370	8.5	20	4	33

Note: The trivial names and spelling are given according to FASS [17]. Q1 is the precursor ion and Q3 the product ion masses. DP, declustering potential; FP, focusing potential; EP, entrance potential; CEP, collision cell entrance potential; CXP, collision cell exit potential; CE, collision energy.

area on flat surfaces [15,16]. In case of collecting a wipe sample from a non-flat surface, the size of the area had to be carefully measured after sampling.

The tissue was taken out from the package and cut in half. One half was used for collecting the wipe sample, and the other half to clean the plastic frame after sampling to avoid cross-contamination between samples. A special wipe pattern was employed to collect the sample [15,16]. The tissue part with the collected sample was folded and placed into a screw-capped test tube and stored in freezer prior to analysis.

Sample Preparation Procedure

After the samples were thawed, the wipe tissue was pushed to the bottom of the tube using disposable stick. Then, 5 mL ethanol and 100 μL of each internal standard solution were added to the tubes, and the samples were shaken for 60 min (IKA-Vibrax WXR, Labassco, Stockholm). A 1.5-mL aliquot of the sample solutions was withdrawn and transferred to micro-vials and evaporated almost to dryness in a Speed Vac Concentrator (Savant Instruments Inc., Farmingdale, US) to avoid decreased recovery, possibly due to analyte decomposition or volatilization. The samples were then re-dissolved in 100 μL 5% methanol in water and transferred to HPLC vials after 30 min.

The samples were then analyzed using HPLC-MS/MS. Quantitative determination of the 12 selected antibiotic substances in the samples was obtained using external standards and could be achieved in one run of each sample. The internal standard was used to verify the sample preparation procedure. Quantification was achieved based on the relative response ratio of the MS/MS daughter ion signal between the external standard and each analyte.

Validation of the Screening Method

A large number of antibiotic substances were considered for the screening method. A survey was used to document frequently used antibiotics [18]. FASS [17] was used to identify the active substances in these drugs. The investigated substances are listed in Table 1. The aim was to find an analytical method where a large number of antibiotic substances could be adequately determined in the same analysis. Previous studies of spill and leakage of cytostatics [1] have shown that a surface detection limit (SDL) at least in the level of 0.05 ng/cm^2 would be desired for a screening method.

For a method intended for supervisory screening, other performance criteria, such as simplicity, robustness, and high sampling efficiency, also have high priority. This means that the same analytical performance, as for regular analytical methods, cannot be expected to be obtained. The primary criteria to validate the method as useful were to determine as many substances as possible in the same analysis and at a SDL at 0.05 ng/cm^2 or lower. The described screening method has been validated in this context.

The analytical method was validated using spiked samples. Unused wet tissues (cut in half) were placed in screw-capped test tubes. A 100 μL aliquot of a standard solution mixture with different antibiotic substances corresponding to

40–300 ng/sample was pipetted into each test tube using a micropipette (Finnpipette, Labsystems, Helsinki, Finland). The test samples were then treated according to the sample pre-treatment procedure prior to analysis. The substances that could be analyzed simultaneously with adequate chromatography, MS-resolution, and with the desired detection limit were selected for further validation of the screening method.

The sampling efficiency from the surface materials was also investigated. A mixed standard solution with all selected substances was prepared in methanol. This solution was spiked onto each surface material in triplicate. Each spiking, 100 μL, was pipetted using a micropipette (Finnpipette, Labsystems, Helsinki, Finland) to 100 cm^2 areas marked up on each surface material. The spikings were left until the surfaces were visibly dried. Wipe samples were then collected from each spiked area using a wipe tissue and a plastic frame [15,16] to wipe a reproducible surface area size. The test wipe samples were then treated according to the sample pre-treatment procedure prior to analysis. The spiking experiment was repeated at a later occasion, giving two sets of triplicate spiking at surface concentrations in the range of 0.4 – 3 ng/cm^2 for the different substances.

Results and Discussions

HPLC-MS/MS Analysis

The previously developed HPLC-MS/MS method for determination of CP [11] was investigated for the analysis of various antibiotic substances. A similar method has also been presented for determination of antibiotics in sewage waters [7]. Reversed phase HPLC coupled with mass spectrometry has frequently been used for determination of many antibiotic substances [7,8]. It was therefore considered to be efficient to employ reversed phase HPLC for the separation of antibiotic substances.

In the separation of complex mixtures of substances, the use of MS/MS for determination of the substances increases the selectivity as well as the specificity for the individual substances. Many interferences from the sample matrix can be eliminated and improve the method performance. In the optimization of the MS/MS determination, focus was put on the possibility of determining as many substances as possible in one analysis with adequate sensitivity. The initial MS/MS scan parameters were used to analyze the first nine substances Cefadroxil (m/z 364 and 208), Diaveridin (m/z 261 and 245), Trimetoprim (m/z 291 and 230), Cefalexin (m/z 348 and 158), Ofloxacin (m/z 362 and 318), Norfloxacin (m/z 320 and 276), Metronidazol (m/z 172 and 154), Ciprofloxacin (m/z 332 and 288), and Enrofloxacin (m/z 360 and 316). At 7 min and 45 s, the scan parameters were changed to analyze the remaining three substances Demeclocyklin HCl (m/z 465 and 448), Doxycyklin (m/z 445 and 428), and Flukonazol (m/z 307 and 220). This change of scan parameters increased the sensitivity of all substances.

A mixed external standard solution was used for quantification. Two isotope-labeled substances, Enrofloxacin-D5 and Flukonazol-D4, were initially

used as internal standards to verify the quantification of the detected substances in the samples as initial tests showed no decomposition of these compounds. However, later experiments showed that some decomposition of the Enrofloxacin-D5 could occur and lead to falsely high recoveries and an increased variability when using that internal standard for quantification. No significant decomposition was found for Flukonazol-D4. Flukonazol-D4 was therefore used in all calculations although both internal standards were added.

Selection of Antibiotic Substances

Based on the antibiotic substances identified in a survey [18], carried out at Swedish hospitals, 27 antibiotic substances were considered for the screening method. To evaluate the possibility for simultaneous HPLC-MS/MS determination of the selected antibiotic substances, test tubes were spiked with a standard solution of the substances.

Based on this evaluation, a number of the investigated substances had to be excluded. The major reasons for exclusion of substances were either poor chromatography, with inadequate separation, or poor peak shapes, resulting in too low sensitivity. Unsatisfactory mass spectrometric resolution was another reason for exclusion. The excluded substances were mainly different penicillins (e.g., Amoxicillin, Benzylpenicillin, and Cloaxcillin), as well as Ceftadizim, Cefuroxim, and Vancomycin. In three instances (Cefotaxim, Imipenem, and Meropenem), although possible to analyze, the analysis did not give a low enough quantification level for the desired performance of the screening method.

Twelve substances (see Table 3) were found to meet the basic criteria, i.e., be possible to analyze in the same run at an analytical detection limit (ADL)

TABLE 3—*Analytical recovery of the selected substances.*

Substance	Spike (ng)	Recovery (%)	RSD (%)
Cefadroxil	179	95	9
Cefalexin	292	96	5
Ciprofloxacin	43	99	43
Demeclocyklin HCl	131	42	27
Diaveridin	89	80	13
Doxycyklin	133	62	29
Enrofloxacin	50	75	26
Flukonazol	39	93	2
Metronidazol	102	102	6
Norfloxacin	57	105	34
Ofloxacin	58	94	25
Trimetoprim	120	70	17

Note: %, mean recovery of four test samples; RSD, relative standard deviation. The trivial names are given according to FASS [17].

adequate for screening with wipe samples from 100 cm^2 surfaces. All substances, except Demeclocyklin HCl and Doxycyklin, showed recoveries above 70%. The relative standard deviation (RSD) was below 30% for all substances except Ciprofloxacin and Norfloxacin. The recovery tests were carried out at levels within two orders of magnitude above the ADL. This performance was considered adequate for a screening method.

Figure 2 shows a chromatogram with all 12 substances included in the screening method. All substances could be separated within a 12 min chromatographic run. In the chromatogram, based on the total ion current, only ten peaks appear. Metronidazol, Norfloxacin, and Ofloxacin were found to have the same retention time. These substances have, however, different precursor and daughter ions (see Table 2) and could efficiently be resolved via MS/MS fragmentation and be quantified individually although having the same retention time.

Detection Limits

The ADL, defined as three times the background noise, for the selected substances and the surface detection limits (SDL), based on a 100 cm^2 wipe sample, are listed in Table 4. The ADL for the substances in the screening method ranged from 0.3 to 3 ng/sample. The lowest ADL was obtained for Norfloxacin and the highest for Metronidazol. The SDL, expressed as ng/cm^2, will, however, vary depending on the area size of the wiped surface, i.e., the larger size of wiped

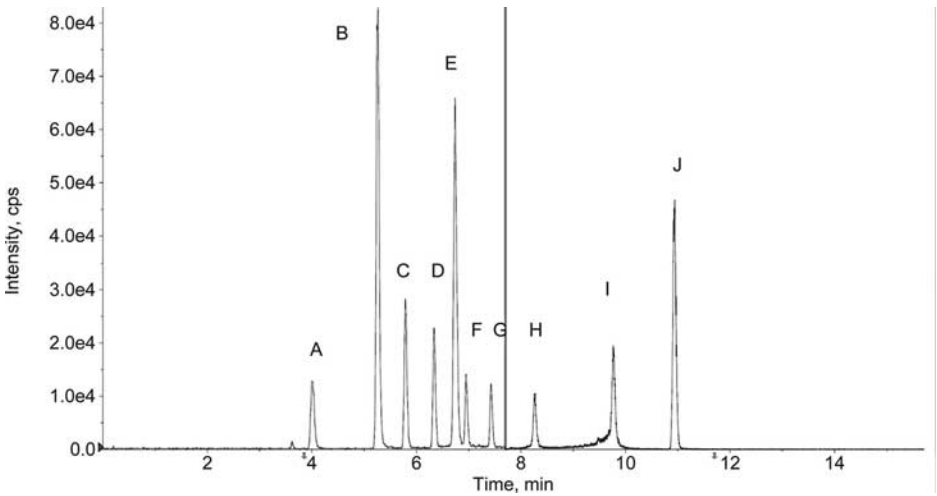

FIG. 2—*Chromatogram of all twelve substances. The chromatogram shows the total ion current (TIC) of all masses. The substances are A-Cefadroxil; B-Diaveridin; C-Trimetoprim; D-Cefalexin; E-Ofloxacin, Norfloxacin and Metronidazol; F-Ciprofloxacin; G-Enrofloxacin; H-Demeklocyklin HCl; I-Doxycyklin; J-Flukonazol. The solid line between peak G and H marks a change in scan parameters (see the text for details).*

TABLE 4—*Analytical detection limits (ADL) and surface detection limit (SDL), based on a 100 cm^2 wipe sample, for the antibiotic substances determined in the screening method.*

Substance	ADL (ng/sample)	SDL(ng/cm^2)
Cefadroxil	0.50	0.005
Cefalexin	3.00	0.03
Ciprofloxacin	0.50	0.005
Demeclocykline HCl	2.00	0.02
Diaveridin	1.00	0.01
Doxycyklin	2.00	0.02
Enrofloxacin	0.30	0.003
Flukonazol	0.70	0.007
Metronidazol	3.00	0.03
Norfloxacin	0.30	0.003
Ofloxacin	0.40	0.004
Trimetoprim	2.00	0.02

Note: The trivial names are given according to FASS [17].

surface area the lower SDL can be obtained. With a surface area size of 100 cm^2, the obtained SDLs were fully satisfactory for the desired performance of the screening method.

Validation of the Screening Method

This method is intended for supervisory screening and other performance criteria, such as simplicity, robustness, and high sampling efficiency, also have high priority besides the analytical performance. In judging the method utility focus has been put on the number of substances that can be analyzed simultaneously at an SDL corresponding to a surface level of 0.05 ng/cm^2 or lower. The described method has been validated in this context.

The recoveries and reproducibility of wipe sampling antibiotic substances from various commonly occurring surface materials were investigated. Surfaces of glass, stainless steel, plastic floor carpet, and laminate bench top were spiked with known amounts of antibiotic substances in the range of 0.4 – 3 ng/cm^2. In this experiment, all spikings were made in triplicate and left to dry before the wipe samples were collected. The spiking experiment was also repeated on a later occasion, and, thus resulting in two set of triple replicates for each surface material.

The result of the recovery and reproducibility study is presented in Table 5. The mean relative recovery was calculated from all six spikings made at two different occasions on each surface material. Considering that this is intended as a screening method, good performance was obtained for Cefadroxil, Cefalexin, Flukonazol, and Metronidazol on all four surface materials and for Diaveridin and Trimetoprim on three surface materials (glass, stainless steel, and bench top laminate). Acceptable performance was obtained for Enrofloxacin,

TABLE 5—*Recovery of antibiotic substances from different surface materials.*

		Surface material							
		Bench		Glass		Plastic floor carpet		Stainless steel	
Substance	Spike ng/cm^2	Rec	RSD	Rec	RSD	Rec	RSD	Rec	RSD
Cefadroxil	1.8	71	12	90	17	77	13	87	16
Cefalexin	2.9	94	17	118	12	77	5	122	10
Ciprofloxacin	0.4	50	42	59	32	19	66	37	35
Demeclocyklin HCl	1.3	6	86	5	65	3	62	5	100
Diaveridin	0.9	87	12	144	19	34	12	149	9
Doxycyklin	1.3	14	34	10	101	6	35	6	36
Enrofloxacin	0.5	30	38	53	20	17	35	37	24
Flukonazol	0.4	84	3	98	6	81	9	99	5
Metronidazol	1.0	101	23	103	16	78	12	110	9
Norfloxacin	0.6	48	61	52	26	34	24	32	36
Ofloxacin	0.6	39	30	56	26	19	21	42	19
Trimetoprim	1.2	67	9	106	21	31	6	113	12

Note: Mean recovery (%) of six samples for each surface material with relative standard deviation (%). Rec, recovery; RSD, relative standard deviation. The trivial names are given according to FASS [17].

Norfloxacin on all surface materials, for Ciprofloxacin, Ofloxacin on three surface materials (glass, stainless steel, and bench top laminate), and for Diaveridin and Trimetoprim on one surface material (plastic floor carpet). Although not fully acceptable, useful performance for a screening method was obtained for Ciprofloxacin and Ofloxacin on one surface material (plastic floor carpet). For Doxycyklin, a useful screening performance was obtained only for two surface materials (bench top laminate and glass) but not for the other surface materials. Finally, it can be questioned if the performance for Demeclocyklin HCl can be regarded as useful for any surface material.

For all substances, the lowest recoveries were obtained from the plastic floor carpet. Many plastic materials show sorption properties [19,20]. These properties will increase the adhesion of organic compounds to the material and may explain the lower recoveries for wipe sampling on plastic floor carpet. This effect may also have a negative impact on the cleaning efficiency of floors covered with plastic carpets.

For practical use of a screening method, in order to get a reasonable overview and be able to compare the level of contamination, it can be appropriate to classify the samples into different categories. Each sample can be given a score from the results according to the following parameters: (i) the number of found substances (one point for each substance) and (ii) the level of the substances. A suitable scoring for each substance is a level of not detected to 0.1 ng/cm^2 = 1 point; 0.1–0.5 ng/cm^2 = 2 points; 0.5–1.0 ng/cm^2 = 3 points; 1–2 ng/cm^2 = 4; 2–5

ng/cm^2 = 5; 5–10 ng/cm^2 = 6; > 10 ng/cm^2 = 7. A high score, thus, means a large contamination. The range of points from the lowest to the highest scored sample can then be divided into four ranges of equal numerical size, representing the four categories: *Low, Medium, High*, and *Very high* level of contamination.

Based on the individual score, each sample can then be classified into one of the contamination categories. If several samples are collected at the same site, the scores can be combined for classification of the contamination level at the site. This classification model, where the results are grouped into concentration ranges, will also contribute to decrease the effects of any deficiencies in the recovery performance for some of the substances and surface material. For example, if the true level of a substance is 0.4 ng/cm^2 and the recovery is 60%, the analysis will show 0.24 ng/cm^2. Both these values will fall into the same group and get the same score. However, if the true level of the substance is 0.15 ng/cm^2 the analysis will show 0.09 ng/cm^2. Here, the analytical result will fall into the lower group, and the sample would be scored too low. Only samples, whose true values are close to the lower border of a group, would risk to be scored too low due to deficiency in analytical performance.

Conclusions

The developed HPLC-MS/MS method has been proven to be suitable for analysis of the 12 selected antibiotic substances in solutions from extracted wipe samples, collected from frequently occurring surface materials. All substances can be separated and quantified simultaneously in one analysis. The obtained ADLs were satisfactory for all 12 selected substances and, based on a 100 cm^2 wipe sample, SDLs well below the desired level were obtained for all substances. For screening investigations using collections of wipe samples, a good or acceptable performance was obtained for ten substances on most surface materials. Although not fully acceptable, useful performance for a screening method was obtained for almost all substances on all surface materials. For comparison of the screening results, a classification procedure can be employed, where each sample gets a score depending on the number of substances found the concentration level in the sample. The samples can then be divided into four groups representing increasing contamination. Such classification simplifies comparisons and will also circumvent any deficiency of recovery performance for some substances and surface materials.

Acknowledgments

Arbetslivets Försäkringsaktiebolag (AFA) [Work Life Insurance Inc.] is acknowledged for the financial support that made this study possible.

References

[1] NIOSH, "Occupational Exposure to Antineoplastic Agents," *Workplace Safety & Health Topics, National Institute for Occupational Safety and Health*, Cincinnati, US,

2010, http://www.cdc.gov/niosh/topics/antineoplastic/default.html (Last accessed Oct. 20, 2010).

[2] CSHP, *Guidelines for the Handling and Disposal of Hazardous Pharmaceuticals (Including Anti-cancer Drugs),* Canadian Society of Hospital Pharmacists, Ottawa, 1993.

[3] American Society of Health-System Pharmacists, "ASHP Guidelines on Handling Hazardous Drugs," *Am. J. Health-Syst. Pharm.*, Vol. 63, 2006, pp. 1172–1193.

[4] HSE, "Safe Handling of Cytotoxic Drugs," *Information Sheet MISC 615, Health and Safety Executive,* Sudbury, Suffolk CO10 2WA, UK, 2003, http://www.hse.gov.uk/pubns/misc615.pdf (Last accessed Oct. 20, 2010).

[5] Work Health Authority, "Cytostatics and Other Drugs with Persistent Toxic Effects," *Ordinance AFS 2005:5,* Swedish Work Health Authority, Stockholm, Sweden, 2005.

[6] Apotekens Service, *Försäljningsstatistik [Sale statistics],* 2010 (in Swe), http://www.apotekensservice.se/Om_statistik/ (Last accessed 2010-10-29).

[7] Lindberg, R., 2006, "Determination of Antibiotics in the Swedish Environment with Emphasis on Sewage Treatment Plants," *Dissertation ISBN 91-7264-045-6,* Umeå University, Umeå, Sweden.

[8] Niessen, W. M. A., "Analysis of Antibiotics by Liquid Chromatography-Mass Spectrometry," *J. Chromatogr.* A, Vol. 812, 1998, pp. 53–75.

[9] Sessink, P. J. M., Anzion, R. B., Van den Broek, P. H. H., and Bos, R. P., "Detection of Contamination with Antineoplastic Agents in a Hospital Pharmacy Department," *Pharm. Weekbl. Sci.*, Vol. 14, 1992, pp. 16–22.

[10] Tuerk, J., Reinders, M., Dreyer, D., Kiffmeyer, T. K., Schmidt, K. G., and Kuss, H.-M., "Analysis of Antibiotics in Urine and Wipe Samples from Environmental and Biological Monitoring – Comparison of HPLC with UV-, single MS- and tandem MS-detection," *J. Chromatogr. B*, Vol. 831, 2006, pp. 72–80.

[11] Hedmer, M., Jönsson, B. A., and Nygren, O., "Development and Validation of Methods for Environmental Monitoring of Cyclophosphamide in Workplaces," *J. Environ. Monit.*, Vol. 6, 2004, pp. 979–984.

[12] Minoia, C., Turci, R., Sottani, C., Schiavi, A., Perbellini, L., Angeleri, S., Draicchio, F., and Apostoli, P., "Application of High Performance Liquid Chromatography/Tandem Mass Spectrometry in the Environmental and Biological Monitoring of Health Care Personnel Occupationally Exposed to Cyclophosphamide and Ifosfamide," *Rapid Commun. Mass Spectrom.*, Vol. 12, 1998, pp. 1485–1493.

[13] Nygren, O. and Lundgren, C., "Determination of Platinum in Workroom Air and in Blood and Urine from Nursing Staff Working with Patients Receiving Cisplatin Chemotherapy," *Int. Arch. Occup. Environ. Health*, Vol. 70, 1997, pp. 209–214.

[14] Ensslin, S., Pethran, A., Schierl, R., and Fruhmann, G., "Urinary Platinum in Hospital Personnel Occupationally Exposed to Platinum-Containing Antineoplastic Drugs," *Int. Arch. Occup. Environ. Health*, Vol. 65, 1994, pp. 339–342.

[15] Nygren, O. and Aspman, O., "Validation and Application of Wipe Sampling and Portable XRF Analysis as an On-Site Screening Method for Assessment of Deposited Aerosols on Workplaces," Aust. J. Chem., Vol. 37, 2004, pp. 1021–1028.

[16] Nygren, O., "Wipe Sampling as a Tool for Monitoring Aerosol Deposition in Workplaces," *J. Environ. Monit.*, Vol. 8, 2006, pp.49–52.

[17] FASS, "Farmaceutiska Specialiteter i Sverige [Pharmaceutical Specialites in Sweden]," *Läkemedelsinsdustriföreningen, Stockholm*, 2010 (in Swe), http://www.fass.se (Last accessed Jan. 11, 2010).

[18] Nygren, O. and Lindahl, R., "Screening of Spill and Leakage of Antibiotics in Hospital Wards," *J. ASTM Int.* (accepted).

[19] Berens, A. R., "Transport of Organic Vapors and Liquids in Polyvinyl(chloride)," *Makromol. Chem. Macromol. Symp.*, Vol. 29, 1989, pp. 95–108.
[20] Saquing, J. M., Saquing, C. D., Knappe, D. R. U., and Barlaz, M. A., "Impact of Plastics on Fate and Transport of Organic Contaminants in Landfills," *Environ. Sci. Technol.*, Vol. 44, 2010, pp. 6396–6402.

Reprinted from JAI, Vol. 8, No. 6
doi:10.1520/JAI103453
Available online at www.astm.org/JAI

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Screening of Spill and Leakage of Antibiotics in Hospital Wards

ABSTRACT: This paper presents a two-phase study of spill and leakage of antibiotics in hospitals. The first phase was a screening of spill and leakage at 21 hospital wards in 16 hospitals. Phase two was an extended investigation, where different measures to reduce spill and leakage were implemented and a follow-up screening was made to evaluate the effect of the measures. At the screening, 206 samples were collected. The result was used to classify the wards into four classes: *Low*, *Mean*, *High,* and *Very high*. Spatial distribution patterns and the effect of compounding systems were also investigated. The screening showed that spill and leakage occur at all wards. Eleven of the 21 wards had *High* or *Very high* contamination level. This result also showed that the substances were distributed according to three possible patterns. The compounding systems also had an impact on the spill and leakage. All four wards that used a closed system were found among the six wards with the lowest spill and leakage, while all three wards that used open venting systems were found among the six wards with the highest spill and leakage. The result also showed that it is possible to handle antibiotics with only insignificant spill and leakage, i.e., by using closed systems. Three wards, classified as *Very high*, were included in the second phase. Measures to decrease spill and leakage and reduce the distribution the substances were implemented. After two month, a follow-up screening was carried out. The result showed lower contamination levels at all three wards and the implemented measures had some effect. Simple and easy-to-do measures can contribute to reduce the spill and leakage that occur. There is still, however, a need to discuss

Manuscript received October 6, 2010; accepted for publication April 29, 2011; published online July 2011.

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Cite as: Nygren, O. and Lindahl, R., "Screening of Spill and Leakage of Antibiotics in Hospital Wards," *J. ASTM Intl.*, Vol. 8, No. 6. doi:10.1520/JAI103453.

how to handle antibiotics in a safe way to reduce possible spill and leakage and to prevent the distribution of this spill and leakage.

KEYWORDS: antibiotics, compounding, hospital wards, leakage, occupational exposure, screening, spill, surface contamination, wipe sampling

Introduction

During the past fifteen years, there are many studies published that describe monitoring methods and/or investigations of spill and leakage and/or occupational exposure to antineoplastic drugs. A comprehensive web based database with reference to literature on this topic has been compiled by Dr. Tom Connor at National Institute for Occupational Health in the United States [1]. Although most countries have strict regulations for handling antineoplastic drugs [2–5], many studies [1] show that spill and leakage frequently occur during handling these drugs in hospital wards and in pharmacies. Moreover, the studies show that the staff gets undesired exposure to these drugs.

Antibiotics can be regarded as another heterogenic group of drugs that is frequently used in hospitals. Over 140 times more antibiotics compared with antineoplastic drugs are administered to hospital patients in Sweden [6] and there are only limited regulations for safe handling of antibiotics in medical care, compared with the situation for antineoplastic drugs. It is therefore not unrealistic to assume that the spill and leakage of antibiotics are the same or larger than with antineoplastic drugs.

There are some reviews on analytical methods for antibiotic substances for pharmacokinetic studies and for antibiotic residues in foodstuffs [7,8]. There are also several studies on the distribution of drugs in the environment through sewer effluents [9–12]. Tuerk et al. [13] have compared different analytical methods for determination of antibiotic substances in environmental and biological samples. There are, however, almost no studies on spill and leakage of antibiotics in medical care.

There have been two main purposes of this study. The first aim has been to investigate the spill and leakage of antibiotics in Swedish hospitals using a previously developed and validated screening method [14]. The method is based on wipe sampling and liquid chromatography tandem mass spectrometry (HPLC-MS/MS) for determination of sampled antibiotics. Twelve different antibiotics have been analysed in over 200 samples collected in 21 wards at 16 different hospitals.

The second aim has been to identify measures to reduce the spill and leakage of antibiotic drugs. In a deeper study at three wards, a number of possible preventive measures were identified and suggested to the wards. After some time for implementation, a follow-up screening was carried out and compared with the results from the first screening in order to evaluate the effect of the suggested measures. At the same time an investigation of the cleaning efficiency was also carried out by monitoring the level of antibiotic substances directly before and after cleaning.

Material and Methods

Material and Chemicals

All chemicals were of analytical grade or higher quality, and the water was purified in a Milli-Q water purifier (Millipore Corp., Billerica, MA, US). Twelve antibiotic substances were included in the screening. They are listed below with trivial name and the chemical names in brackets, all according to FASS [15]. The substances were: Cefadroxil [(6R,7R)-7-[(R)-2-Amino-2-(p-hydroxifenyl)acetamido]-3-metyl-8-oxo-5-tia-1-azabicyklo[4.2.0]okt-2-en-2-karboxylsyra], Cefalexin [(6R)-7R-7-[(R)-2-Amino-2-fenylacetamido]-3-metyl-8-oxo-5-tia-1-azabicyklo [4.2.0]okt-2-en-2-karboxylsyra], Ciprofloxacin [1-Cyklopropyl-6-fluoro-1,4-dihydro-4-oxo-7-(1-piperazinyl)-3-kinolinkarboxylsyra], Demeclocyklin HCl [7-Chloro-4.(dimethylamino)-1,4,4a,5,5a,6.11,12a-octahydro-3,6,10.12.12a-pentahydroxy-1,11-dioxo-2-], Diaveridin [2,4-diamino-5-(3,4-dimethoxybenzyl)pyrimidine 5-((3,4-dimethoxyphenyl)methyl)-4-pyrimidinediamine], Doxycyklin [(4S, 4aR,5S,5aR,6R,12aS)-4-Dimetylamino-1,4,4a,5,5a,6,11,12a-oktahydro-3,5, 10,12, 12a-pentahydroxi-6-metyl-1,11-dioxo-2-naftacenkarboxamid], Enrofloxacin [1-Cyklopropyl-7-(4-etyl-1-piperazinyl)-6-fluoro-1,4-dihydro-4-oxo-3-kinolinkarboxylsyra], Fluconazol [2,4-Difluoro-α,α-bis(1H-1,2,4-triazol-1-ylmetyl)bensylalkohol], Metronidazol [1-(2-Hydroxietyl)-2-metyl-5-nitroimidazol], Norfloxacin [1-Etyl-6-fluoro-1,4-dihydro-4-oxo-7-(1-piperazinyl)-3-kinolinkarboxylsyra], Ofloxacin [9-Fluoro-2,3-dihydro-3-metyl-10-(4-metyl-1-piperazinyl)-7-oxo-7H-pyrido [1,2,3-de]-1,4-benzoxazin-6-karboxylsyra], Trimetoprim [2,4-Diamino-5-(3,4,5-trimetoxibensyl)pyrimidin]. As internal standards, the following isotope labelled antibiotics were used: Enrofloxacin-D5, Fluconazol-D4, Norfloxacin-D5. The concentration varied between the compounds but was in the range of 1.5–5.5 μg/ml.

Powder free disposal vinyl gloves (Evercare, Selfatrade AB, Spånga, Sweden) were used when collecting the samples. Wet tissues (Apoliva, Apoteket AB, Stockholm, Sweden) were used for collection of wipe samples. A homemade plastic frame (outer size 14×14 cm and encompassing 10×10 cm $= 100$ cm^2) was used to get a reproducible size of the wipe samples from flat surfaces. Screw capped plastic tubes (Sarstedt, 15 ml, Nümbrecht, Germany) were used to store the wipe samples.

Instrumentation

For the analysis, a PerkinElmer (Norwalk, CT), chromatographic system consisting of two micro-pumps (PerkinElmer series 200) and an auto-sampler (PerkinElmer series 200) was used. The high-performance liquid chromatography (HPLC) was equipped with a YMC Hydrosphere C18 column (YMC Separation Technology Inc., Wilmington, NC, US) 150×4.6 mm id, 5 μm. Acetonitrile in water with 0.1 % of formic acid were used as HPLC-eluent, starting at 15 % of acetonitrile for 2 min followed by a gradient to 70 % after 9 min. The HPLC was coupled to a triple quadrupole mass spectrometer (API 2000 PE Biosystem, Foster City, CA) equipped with an electrospray ion source (TurboIonSpray). The ion spray voltage was set to 5.5 kV (positive mode), and the drying gas was at 350°C.

Sampling Procedure

The sampling for the screening was then made according to a previously reported procedure [16,17]. Wipe samples were collected from suitable surfaces using the plastic frame for flat surfaces. The area of non-flat sampled surfaces was carefully measured after the wipe sample was taken. Powder free disposal gloves were used and changed between each sample. For each sample a wet tissue was used. The tissue was cut in two halves using a pair of scissors. One half of the tissue was used to collect the sample, and the other half was used to clean the plastic frame after sampling. The previously validated wipe procedure was employed when taking the samples. After wiping the surface, the wet tissue was folded once more and then rolled and placed into a screw capped plastic test tube. The samples were then stored in freezer (−20°C) until analysis.

Analytical Procedure

The samples were analysed according to a method described elsewhere [14]. In short, the samples were thawed, and the tissue in each tube was compressed to the bottom. Then, 5 ml of ethanol and 100 μl of each internal standard solution were added, and the samples were shaken for 60 min. Then, 1.5 ml of the solution was transferred to micro-vials and evaporated to almost to dryness in a Speed Vac Concentrator (Savant Instruments Inc., Farmingdale). The samples were re-dissolved in 100 μl of 5 % methanol in water transferred to HPLC vials after 30 min and analysed by HPLC-MS/MS.

Selection of Sites for Screening

For selection of suitable sites for this screening, an inquiry (see Appendix A) was sent out to 64 wards at 42 hospitals in the northern half of Sweden to investigate, which antibiotic drugs that regularly were used and also to identify sites that were prepared to participate in the screening. Of the 64 wards, 36 answered the inquiry of which 24 were prepared to participate and 12 declined. Of these 24 sites, three were not included in the screening in the end due to difficult travel logistics. The screening was, thus, carried out at 21 hospital wards (intensive care, haematology, surgery, or general wards) in 16 different hospitals in Sweden. The hospitals ranged from University hospitals to minor regional hospitals. All hospitals were public.

Screening Procedure

Each ward was visited during the screening. At the visit, before any samples were collected, the facilities, compounding systems, compounding procedures, administration routines as well as cleaning and waste handling routines etc., were recorded and documented. During this evaluation focus were particularly put on the compounding system used, the place for compounding, the cleaning procedure, the cleaning frequency, the presence of written instructions for compounding, the average number of daily compounded doses, the number of

individuals that made drug compounding as well as their experience (no of years with compounding) and the drugs that actually were used at the time of the screening. This documentation was also verified with a nurse at each ward that normally carried out compounding and administration of antibiotic drugs (usually the nurse that was responsible for the drug room).

At each ward, approximately 10 samples were collected. The samples were taken from benches used both for compounding and not for compounding, drug shelves, waste containers, sinks, and the floor in the drug room, and from the toilet seat and the floor in patient toilets and from other relevant locations at the wards (e.g., nurse office, coffee room, etc.,). All sampling was carried out from June to November 2008. Appendix B summarizes the sampling locations at each ward.

From the results, the mean and median values, range and number of samples above the detection limit and the number of sampling locations with identified substance, were calculated for each substance. In the mean value calculation, all results below the analytical detection limit (ADL) were assigned (ND-not detected) to

$$ND = \frac{ADL}{\sqrt{2}}$$

According to Hornung and Reed [18], this method to handle ND-values gives more adequate results than ND = ADL/2 when data has a log-normal distribution. This is normally the case with data series that has a lower limit but virtually no upper limit, like in this case.

Classification Procedure

To get a reasonable overview and be able to compare the level of spill and leakage, the wards were classified into four categories. Each ward was given a score according to the results of the following parameters: (i) number of substances found (one point for each substance), (ii) the number of samples with substances (one point for each substance in a sample), and (iii) the level of the substances (for each substance $0.01-0.1$ ng/cm$^2 = 1$ point; $0.1-0.5$ ng/cm$^2 = 2$ points; $0.5-1.0$ ng/cm$^2 = 3$ points; 1–2 ng/cm$^2 = 4$; 2–5 ng/cm$^2 = 5$; 5–10 ng/cm$^2 = 6$; >10 ng/cm$^2 = 7$). A high score, thus, meant a large spill and leakage. The range of points from the lowest to the highest score was divided into four ranges of equal numerical size, representing the four categories: *Low, Medium, High,* and *Very high* level. Based on the individual score, each ward was then classified into one of the categories.

Evaluation of Spatial Distribution Patterns

Based on the results for different sampling locations at each ward the spatial distribution patterns were evaluated. The results from primary surfaces, where antibiotics were handled, i.e., work benches for compounding drugs,

drug shelves, etc., and the results from secondary surfaces, where antibiotics not were handled, i.e., floors, benches not used for compounding, etc., were considered. Based on the differences between the results from primary and secondary surfaces various spatial distribution patterns were identified.

Effect of Compounding System on the Level of Spill and Leakage

Numerous studies on handling of cytostatics have shown that the compounding system have a significant impact on the level of drug spill and leakage [1]. In order to investigate the effect of the compounding system during preparation of antibiotic drugs, the compounding systems used at the wards were classified into three categories: (i) open systems with a venting-needle without filter or with the traditional "milking technique," (ii) some type of spike with filter for venting (e.g., Mini-spike, Braun Medical AB, Danderyd, Sweden or Venting-needle, Baxter Medical AB, Kista, Sweden), and (iii) closed compounding system (e.g., PhaSeal, Carmel Pharma AB, Gothenburg, Sweden or Tevadaptor, Teva Sweden AB, Helsingborg, Sweden). For each ward, the classification level of the ward was compared to the compounding system category used at the ward.

Selection of Sites for the Investigation of Preventive Measures

In the second part of this study, three wards, classified as having *Very high* contamination level, were selected. The compounding and administration procedures were recorded step by step in order to identify possible causes to the spill and leakage. Based on this documentation, a number of preventive measures to minimize spill and leakage, as well as measures to prevent the spatial distribution of emerged spill and leakage were suggested. Examples of suggested measures are presented in Appendix C. The preventive measures were individually suggested to each ward based on their particular situation and presented to the staff in a written document. At each ward, the staff then decided by themselves, which of the suggested measures that should be implemented. About two month after the selected measures were introduced at the ward, a follow-up screening was carried out. At the follow-up screening, wipe samples were collected on the same surfaces as in the first screening.

Assessment of the Cleaning Procedure

The cleaning procedure may have a significant impact on the level of spill and leakage of cytostatics [1,19,20]. During the second part of this project, a possibility to investigate the effect of the generally used cleaning procedure was possible. At the follow-up screening, samples were also collected immediately before and after general cleaning of the work benches and floor in the drug rooms and on the floor in the patient toilets.

The general cleaning procedure was similar at all three wards. The floor was wiped with a mop dampened in a water-detergent solution. The mop cloth was changed between each room, and the used mop clothes were washed in a laundry, dried, and reused, until worn out.

Results and Discussion

Initial Survey

Table 1 shows the antibiotic substances that were identified to be active compounds in the drugs that were specified by the wards in the survey to be regularly used. Twenty-five compounds were identified to be used in at least one drug at one of the wards. Two of the compounds determined in this screening (Cefalexin and Diaveridin) were not listed in the survey as regularly used. A limited number of compounds were present in frequently used drugs. Ciprofloxacin was present in drugs used at all sites, and Cefuroxim, Fluconazol, Meropenem, Metronidazol, and Trimetoprim were present in drugs used at more than 70 % of the screened sites. Eight of the compounds were present in drugs that were used in less than 15 % of the sites. Three compounds (Cefuroxim, Meropenem, and Tazocin) were present in frequently used drugs but were not analysed in the screening. However, the four compounds (Ciprofloxacin, Fluconazol, Metronidazol, and Trimetoprim) that were present in the most frequently used drugs were all included in the screening.

Screening

Twelve different antibiotic substances were analysed in 206 samples collected at 21 wards at 16 different hospitals. The results from the screening are summarized in Table 2.

Ciprofloxacin was the substance that occurred most frequently in the samples and was identified in samples from all wards. It was a substance that was specified in the survey to be present in drugs that were administered both as infusion and as tablets. Ciprofloxacin occurred in at least one sample at >10 ng/sample at all wards. Other substances that were present in amounts >10 ng/sample at many wards were Metronidazol (19 wards), Fluconazol (17 wards), and Trimetoprim (15 wards). All three of these substances occur in antibiotics administered both as infusion and tablets.

Metronidazol had the highest mean value of 2.4 ng/cm^2, the highest median value (0.061 ng/cm^2), as well as the second highest individual value (205 ng/cm^2). Trimetoprim showed the highest individual value, which was 340 ng/cm^2, and the second highest mean value (1.92 ng/cm^2). The second highest median value was 0.022 ng/cm^2 and was obtained for Doxycyklin. Diaveridin was the substance that showed both the lowest mean and median values, which were 0.002 and <0.001 ng/cm^2, respectively.

It is also worth to note that the substances that were specified in the survey to only be present in drugs administered as tablets (Cefadroxil, Cefalexcin, Enrofloxacin, Norfloxacinoch, and Ofloxacin) also could be identified in several samples. At least one of these substances was present at a level >10 ng/sample at eleven wards. The levels of these substances, however, were generally lower compared with substances present in drugs that were administered by infusion too. This means that handling tablets, e.g., splitting tablets, filling tablet dispensers (a box with compartments for several doses, usually for one week), also caused distribution of drug particles. It is, thus, also necessary to consider handling tablets to efficiently reduce spill and leakage of antibiotics.

TABLE 1—*Antibiotic substances used in drugs that are regularly used at the wards according to the survey.*

Hospital no/ward	Amoxicillin	Benzylpenicillin	**Cefadroxil**	**Cefalexin**	Cefotaxim	Ceftadizim	Cefuroxim	**Ciprofloxacin**	Clindamycin	Cloaxillin	**DemeclocyklinHCl**	**Diaveridin**	**Doxycyklin**	**Enrofloxacin**	Flucloaxillin	**Fluconazol**	Gentamicin	Imipenem	Meropenem	**Metronidazol**	**Norfloxacin**	**Ofloxacin**	Pivmecillinam	Tazocin	Tobramycin	**Trimetoprim**	Vancomycin
1/Intensive care							X	X			X		X			X		X	X	X	X	X				X	
1/Surgery			X				X	X		X			X			X				X		X				X	
2/Hematology						X		X			X		X			X					X	X		X		X	
2/Infection		X						X	X	X						X	X		X	X				X		X	
3/Intensive care					X		X	X		X			X			X		X	X	X						X	
4/Surgery							X	X	X				X						X	X	X	X		X			
5/Hematology			X			X	X	X			X		X			X		X	X		X	X		X		X	
6/Hematology-Oncology		X	X				X	X					X	X		X			X	X	X	X		X		X	
7/Infection		X	X		X		X	X		X										X		X		X			
7/Hematology	X		X		X		X	X			X		X		X	X			X	X	X	X				X	X
8/General ward	X	X	X				X	X			X		X			X				X			X			X	
9/Infection		X					X	X		X	X		X	X		X				X	X	X		X	X		
9/Hematology		X					X	X	X		X		X			X			X	X	X	X		X		X	X
10/Infection					X		X	X	X	X						X				X							X
11/Infection		X			X		X	X		X								X		X							
12/Surgery			X				X	X	X										X	X				X			
13/General ward		X			X			X							X		X		X	X				X		X	
14/Hematology			X					X			X		X			X				X	X	X				X	
14/Infection		X					X	X	X	X			X			X				X	X			X		X	
15/General ward			X		X			X		X						X			X	X	X			X		X	
16/General ward					X		X	X		X						X				X						X	

Note: The compounds in bold have been determined in this screening. Substance trivial names are given according to FASS [15].

TABLE 2—*Results of the screening.*

Compound	Mean, ng/cm^2	Median, ng/cm^2	Range (min–max), ng/cm^2	Number of samples above DL	Number of samples above QL	Number of wards above DL	Comments
Cefadroxil	0.028	0.006	<0.001–0.595	161	73	19	
Cefalexin	0.006	0.004	<0.001–0.070	152	49	19	Second lowest mean and median
Ciprofloxacin	1.702	0.048	<0.001–312.6	203	161	20	Second highest mean and median, most samples above QL
Demeclocyklin HCl	0.037	0.013	<0.001–1.019	199	116	20	
Diaveridin	0.002	<0.001	<0.001–0.077	163	8	18	Lowest mean and median
Doxycyklin	0.073	0.022	<0.001–0.900	204	129	20	Most frequently occurring compound
Enrofloxacin	0.015	0.007	<0.001–0.152	192	80	19	
Fluconazol	0.581	0.009	<0.001–7.606	202	100	19	
Metronidazol	2.407	0.061	<0.001–2.052	202	154	20	Highest mean and median, 2nd highest concentration
Norfloxacin	0.041	0.016	<0.001–0.550	178	118	19	
Ofloxacin	0.031	0.011	<0.001–0.797	202	107	19	
Trimetoprim	1.927	0.007	<0.001–339.6	181	94	20	Highest concentration

Note: A total of 206 samples collected from 21 wards at 16 different hospitals. DL–detection limit; QL–quantification limit (10 × DL). Substance trivial name according to FASS [15].

Both Cefalexcin and Diaveridin were discovered in several samples, although no ward had specified that drugs, containing these substances were used. One reason to the occurrence of these substances could be that the staff was unaware of that such drugs were used or that such drugs had been used before or after the time of the survey.

Classification of the Wards

To be able to get an overview of all data and to compare the wards, a classification system was designed. Three basic parameters were considered in the classification: (i) the number of substances found, (ii) the number of contaminated locations, and (iii) the level of the spill. The classification levels are relative to the range from the lowest to the highest score for each parameter. The reason for a relative scale was that there was no information on which level of spill is to be expected. With a relative scale, additional data can be added in the future to adjust the scale.

Table 3 shows the classification of the wards that participated in the screening. Of the 21 wards, four were classified as *Low*, six as *Medium*, eight as *High* and, finally, three as *Very high*. This means that eleven of the 21 wards (52 %) were classified as *High* or *Very high*.

At the wards with the highest classification level, at least one substance was found in all samples. A total of nine different substances were found of which three substances occurred at levels >5 ng/cm^2. This indicates a significant spill and leakage at these wards with a significant distribution to secondary surfaces.

At the wards with the lowest classification, substances were found in, at the most, 1–4 samples. Not more than two to four substances were found, all at levels below 0.5 ng/cm^2 and most substances < 0.1 ng/cm^2. This result shows that it is possible to handle antibiotics with only insignificant spill and leakage. The screening, however, showed that only 14 % of the wards (three of 21) manage to handle antibiotics in that way.

Spatial Distribution Patterns

Based on the data from all sampling location at all wards, considering the distribution from primary surfaces towards secondary surfaces, three different distribution patterns could be perceived.

Pattern 1: Small spill and leakage occurred on primary surfaces at the compounding place like work benches and waste containers. There was no or only insignificant distribution to secondary surfaces such as benches, where no drugs were handled or to the floor in the drug room or to other rooms. This pattern indicates that the way of working is well-adapted for the purpose. Only limited spill and leakage occur. The routines for cleaning are also suitable and prevent that emerging spill and leakage will be distributed to secondary surfaces.

Pattern 2: Larger spill and leakage can be shown on primary surfaces at the compounding place such as work benches and waste containers but also a significant distribution to secondary surfaces occur, e.g., to adjacent benches, where no drugs are handled, or to the floor. Drugs can usually also be found in patient toilets and wash rooms. This pattern indicates that spill and leakage

TABLE 3—*Classification of the contamination level at the wards that participated in the screening.*

Hospital	Ward	Class
1	Intensive care	+
1	Surgery	++
2	Hematology	+
2	Infection	+
3	Surgery	0
4	Intensive care	0
5	Hematology	0
6	Hematology/oncology	+
7	Infection	−
7	Hematology	+
8	General care	+
9	Infection	0
9	Hematology	+
10	Infection	0
11	Infection	−
12	Surgery	−
13	General care	−
14	Hematology	++
14	Infection	0
15	General care	++
16	General care	+

Note: Class levels are: Low (−), Medium (0), High (+), and Very high (++). The classification criteria are described in the text.

frequently occur. Moreover, the routines for cleaning are not fully sufficient to prevent that spill and leakage are distributed to secondary surfaces.

Pattern 3: Large spill and leakage occur and are distributed on to both primary and secondary surfaces. Frequently, there are larger levels of contamination on the secondary surfaces such as the floor, the sink in wash rooms and patient toilets. The occurrence of this distribution pattern indicates that the way of working regularly results in spill and leakage. Furthermore, the result shows that the routines for cleaning are not sufficient to prevent that the spill and leakage is significantly distributed to secondary surfaces.

All the wards that had been classified as *Low* showed distribution pattern 1. The wards that had been classified as *Very high*, all showed distribution pattern 3, as well as most of the wards that had been classified as *High*. A majority of the wards that had been classified as *Medium* showed distribution pattern 2.

Effect of the Compounding System

Many antibiotics aimed for infusion are sold as dry substance in glass vials sealed with an aluminum cap and a rubber stopper. For illustration, a short

description of how an infusion bag can be prepared and at which operations spill and leakage can be expected to emerge is given below.

When compounding antibiotics for infusion, a disposal syringe is filled with a suitable aliquot of saline solution from a storage bottle. The syringe is fitted with an injection needle and the liquid is injected into the vial through the rubber stopper. To eliminate the increased pressure that builds up during the injection the traditional milking technique or some kind of venting system are used, e.g., an extra open injection needle, a filter spike (e.g., Braun Mini-spike or Baxter Venting-needle) or a closed system (e.g., PhaSeal or Tevadaptor). During this operation, dry substance or liquid may be expelled to the air as aerosol depending on how efficient the venting system can collect the formed aerosol. The dry substance is then dissolved in the vial, and the desired volume of the dissolved drug is drawn back into the syringe. Then, the syringe is disconnected from the drug vial. Liquid spill or aerosol emission can occur at this operation. The syringe is then connected to an inlet port of the infusion system, and the drug is injected into the infusion liquid. In some occasions, the syringe is disconnected from the inlet port after injection and in other occasions, the syringe is left connected to the inlet port. In the first case, spill or aerosol emission can occur when the syringe is disconnected. Other operations when spill or aerosol emission can occur are when the tubing in the infusion system is filled, when drug infusion bag is connected to or disconnected from the infusion system.

Of the wards participating in the screening, four wards stated that they used some kind of closed system (e.g., PhaSeal or Tevadaptor) for compounding antibiotics for infusion. Three wards stated that they regularly used only an extra open injection needle for venting the vials or milking technique. The other 14 wards used some kind of injection needle with filter or spike (e.g., Braun Mini-spike or Baxter Venting-needle) for venting the vials.

A comparison of the classification results and the compounding system used at the wards showed a clear correlation. Figure 1 shows the result of this comparison. All four wards that used closed systems were among the six wards that had the lowest spill and leakage, i.e., classification *Low* or *Medium*. Correspondingly, all three wards that used milking technique or an open injection needle for venting were among the six wards with the highest contamination level in the screening, i.e., classification *High* or *Very high*.

Description of Preventive Measures

Three wards, with classification *Very high,* were selected for the extended investigation to study the effect of preventive measures. All three wards were visited twice during the extended study. At the first visit, the compounding work was studied operation for operation and the whole compounding procedure was documented. At a second visit a follow-up screening was carried out.

After examination of the compounding procedures a range of measures to prevent spill and leakage as well as the distribution of any emerging spill were documented. Below are examples of preventive measures that were suggested (see also Appendix C).

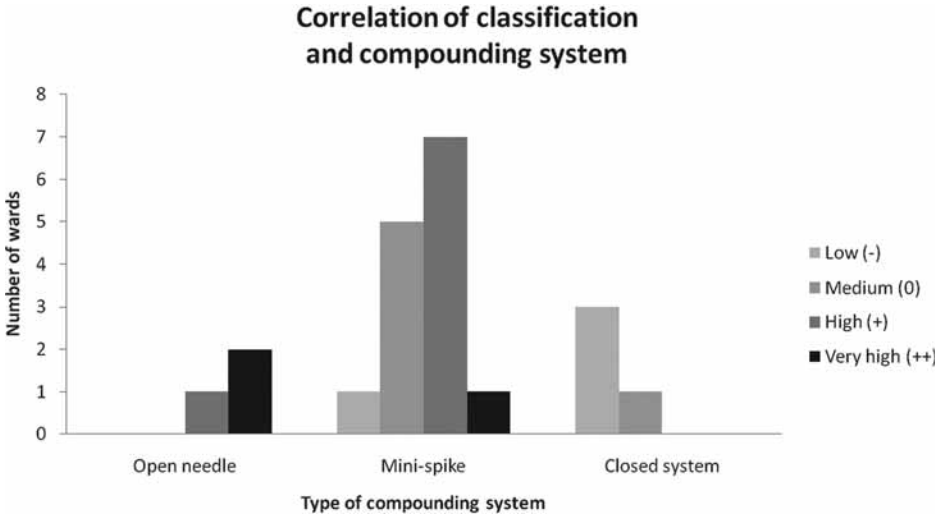

FIG. 1—*Correlation of classification of wards and the type of compounding system used for antibiotics. The classification levels (Low, Medium, High, and Very high) are described in the text.*

Measures to Minimize Spill and Leakage During Compounding—Consider using a closed system for compounding antibiotics. Studies of cytostatics [1] have shown that using closed compounding systems can contribute to significantly reduce the spill and leakage.

When filling tubings in infusion systems, collect any emerging liquid from the tubing nozzle over a bench cover with plastic bottom or a collection container. Do not hold the tubing nozzle so any spill land on the floor or bench top.

When using infusion bags pre-filled with antibiotics, fill the tubings of the infusion system first with saline solution to evacuate air instead of using the drug solution and administer saline solution after infusion is completed to evacuate any remaining drug solution before disconnecting the infusion system from the patient. With this procedure, any emerging spill during these operations will contain only saline solution and minimize the risk for drug spill.

When handling tablets, e.g., splitting tablets and filling drug dispensers, use disposal gloves and carry out these operations on a bench cover with plastic bottom to collect any emerging drug dust. Discard the cover and gloves after the operations are finished.

Prepare infusion systems as much as possible in the drug room. This minimize the risk for spill and leakage in the ward rooms.

Measures to Minimize Distribution of Emerging Spill and Leakage—Use disposal gloves when doing compounding and change gloves between each compounding. This prevents skin exposure but also to prevent that any spill on the gloves are distributed from one compounded infusion bag to the next.

Carry out compounding on a bench cover with plastic bottom that is discarded after each compounding. Any emerging spill will then be collected on the cover and this minimizes the risk that the spill becomes distributed to other surfaces.

Change disposal gloves when leaving the drug room with the prepared drug. This minimize the risk to distribute drugs from contaminated gloves.

Any visible spill on benches, floor or other surfaces should immediately be wiped and the surface should then be cleaned with water and detergent before any disinfection with 70 % alcohol. Alternatively, use a cleaning alcohol solution, e.g., 45 % alcohol in water with a tenside (e.g., M-Ytdes 45+, Kemetyl, Stockholm). Drug spill that is allowed to dry will be more difficult to clean [1].

Always clean benches and other surfaces with water and detergent before any disinfection with 70 % alcohol solution. Alternatively, use a cleaning alcohol solution (e.g., M-Ytdes 45+). Drug contaminations are generally more soluble in water than alcohol and will be easier to wash away with water [1]. If a cleaning alcohol solution is used on a daily basis for general cleaning, a thin film of tenside will be formed. It is therefore recommended to clean the surfaces with water on regular bases to remove this film, e.g., once a week.

Another parameter to reduce the distribution of spill and leakage is an efficient and well-adapted general cleaning procedure. Studies have shown that there are significant difficulties to wash away drug spill and leakage [1,19,20]. If spill is taken care of immediately, it is much easier to wash away. If the spill has been allowed to dry, it is much more difficult to clean [1].

Patients under treatment usually excrete significant amounts of drugs and drug contaminated surfaces are often found in patient areas. Particularly, patient toilets have been shown to be highly contaminated [19].

When cleaning the floor in areas where drug spill can be expected, it is recommended to mop the floor two times with a change of mop cloth in between to improve the result and contribute to decrease the distribution of emerged spill.

Currently, cleaning within medical care in Sweden is much focused on aseptic procedures to prevent growth and distribution of bacteria and other germs [21]. Good information on how to wash away drug spill is, however, often lacking. It is essential that the cleaning staff gets correct information and education to carry out an adequate cleaning.

Benefits of Preventive Measures

To evaluate the effect of the suggested measures, three wards were invited to participate in a limited study. At each ward, the staff got a list of suggested measures and decided among themselves, which of the suggested measures they should implement. About two month after the implementation of the selected measures, a follow-up screening was carried out. At this screening wipe samples were collected at the same locations as in the first screening. The results from this follow-up screening are reported in Tables 4–6.

Hospital 1, Surgery ward, was classified as *High*, which was a reduction from *Very high* in the first screening. There was at least one antibiotic substance in all samples (see Table 4). Ten different substances were found compared

TABLE 4—*Result from the follow-up screening at Hospital 1, Surgery ward. Substance trivial names according to FASS [15].*

Substance	Place	Level, ng/cm^2						
		0,01–0,1	0,1 – 0,5	0,5 – 1,0	1,0 – 2,0	2,0 – 5,0	5,0 – 10,0	> 0,0
Cefadroxil	Drug room, floor, below compounding bench, after cleaning	X						
Ciprofloxacin	Drug room, sink at compounding bench, in stainless steel	X						
	Drug room, floor, below compounding bench, after cleaning	X						
	Patient toilet, floor, below toilet, after cleaning	X						
Demeclocyklin HCl	Drug room, compounding bench in stainless steel	X						
	Drug room, work bench, below drug shelves	X						
	Drug room, floor, below compounding bench, before cleaning	X						
	Wash room, sink for urine measurements, dirty side	X						
	Drug room, work bench, below drug shelves	X						
	Drug room, floor, below compounding bench, before cleaning	X						
	Drug room, drug shelf (trimetoprim)	X						
Flukonazol	Drug room, work bench, below drug shelves	X						
	Drug room, floor, below compounding bench, before cleaning	X						
	Drug room, floor, below compounding bench, after cleaning	X						
Metronidazol	Drug room, work bench, below drug shelves		X					
	Drug room, floor, below compounding bench, before cleaning			X				
	Wash room, floor, below waste box		X					
	Wash room, floor, below clean side of sink	X						
	Drug room, floor, below compounding bench, after cleaning			X				
	Patient toilet, floor, below toilet, after cleaning		X					
Norfloxacin	Drug room, compounding bench in stainless steel	X						
Ofloxacin	Drug room, compounding bench in stainless steel	X						
Trimetoprim	Drug room, compounding bench in stainless steel		X					

TABLE 4—*Continued*

Substance	Place	Level, ng/cm^2						
		0,01–0,1	0,1 – 0,5	0,5 – 1,0	1,0 – 2,0	2,0 – 5,0	5,0 – 10,0	> 0,0
	Drug room, work bench, below drug shelves	X						
	Drug room, floor, below compounding bench, before cleaning	X						
	Drug room, drug shelf (trimetoprim)		X					
	Drug room, floor, below compounding bench, after cleaning	X						
	Patient toilet, floor, below toilet, after cleaning	X						

with seven at the first screening, which resulted in the classification level *High*. The levels of the substances were considerably lower than in the first screening. In the majority of the samples, the levels were <0.1 ng/cm^2. Only Metronidazol was found at levels >0.5 ng/cm^2 in two samples.

An improvement was also obtained at Hospital 14, Hematology ward. In nine of ten samples, there was at least one substance, which can be seen in Table 5. Only four substances were found compared with six in the first screening. None of the substances were present in levels >0.5 ng/cm^2 in any of the samples. This resulted in a change of classification level from *Very high* to *Medium* in the follow-up screening.

Twelve of the samples at Hospital 15, General ward, showed presence of antibiotic substances at the follow-up screening (see Table 6). Nine different substances were found compared with six in the first screening. A majority of the samples showed levels <0.1 ng/cm^2. The levels were consequently considerably lower than at the first screening. The ward was, however, classified as *High*, compared with *Very high* at the first screening. The major reason for this was the large number of substances present.

The results from the follow-up screening (see Tables 4–6) show for all three wards that there were lower amounts of antibiotic substances in the samples compared with the first screening. It were mainly on the surfaces that had high levels in the first screening that showed lower levels in the follow-up screening and this was valid for both primary and secondary surfaces. This means that the spill and leakage that occur had decreased. At the follow-up screening, the wards had implemented several of the suggested measures for two month. At all three wards, disposal gloves were used and changed after each compounding. The compounding was carried out on a bench cover, with plastic bottom, that was changed after each compounding. The bench top surfaces were cleaned and disinfected with cleaning alcohol solution (M-Ytdes45+) instead of 70% alcohol. None of the wards had changed to a closed compounding system. Two of the wards had changed from open venting system to a filter spike for venting.

TABLE 5—*Result from the follow-up screening at Hospital 14, Hematology ward. Substance trivial names according to FASS [15].*

Substance	Place	Level, ng/cm^2						
		0,01–0,1	0,1–0,5	0,5–1,0	1,0–2,0	2,0–5,0	5,0–10	>10
Cefadroxil	Patient toilet, floor, below toilet, after cleaning	X						
Flukonazol	Drug room, compounding bench to the right, before cleaning	X						
	Drug room, floor, below compounding bench, before cleaning		X					
	Drug room, compounding bench to the right, after cleaning	X						
	Drug room, floor, below compounding bench, after cleaning	X						
	Patient toilet, floor, below toilet, before cleaning		X					
	Patient toilet, floor, below toilet, after cleaning		X					
Metronidazol	Drug room, floor, below compounding bench, before cleaning	X						
	Drug room, drug shelf (Metronidazol)	X						
	Drug room, floor, below compounding bench, after cleaning	X						
Trimethoprim	Drug room, compounding bench to the left, before cleaning	X						
	Drug room, compounding bench to the right, before cleaning	X						
	Drug room, floor, below compounding bench, before cleaning		X					
	Drug room, drug shelf (Metronidazol)	X						
	Drug room, compounding bench to the right, after cleaning	X						
	Drug room, floor, below compounding bench, after cleaning	X						
	Patient toilet, floor, below toilet, before cleaning		X					
	Patient toilet, floor, below toilet, after cleaning		X					

TABLE 6—*Result from follow-up screening at Hospital 15, General ward. Substance trivial names according to FASS [15].*

Substance	Place	Level, ng/cm^2						
		0,01–0,1	0,1–0,5	0,5–1,0	1,0–2,0	2,0–5,0	5,0–10	>10
Cefadroxil	Drug room, floor, below bench beside compounding bench, before cleaning	X						
	Drug room, drug shelf (Trimetoprim)	X						
	Drug room, floor, below compounding bench, after cleaning	X						
	Drug room, floor, below bench beside compounding bench, after cleaning	X						
	Patient toilet, floor, below toilet, before cleaning	X						
	Patient toilet, floor, below toilet, after cleaning	X						
	Drug room, compounding bench, after cleaning with M-Ytdes45+	X						
	Drug room, compounding bench, after cleaning with water	X						
	Drug room, bench beside compounding bench, after cleaning with M-Ytdes45+	X						
Cefalexin	Drug room, floor, below bench beside compounding bench, before cleaning	X						
	Drug room, drug shelf (Trimetoprim)	X						
	Drug room, floor, below compounding bench, after cleaning		X					
	Drug room, floor, below bench beside compounding bench, after cleaning	X						
	Patient toilet, floor, below toilet, after cleaning	X						
	Drug room, compounding bench, after cleaning with water	X						
Demeclocycline HCL	Drug room, bench beside compounding bench, before cleaning	X						
	Drug room, sink, before cleaning	X						

TABLE 6—*Continued*

Substance	Place	Level, ng/cm^2						
		0,01–0,1	0,1–0,5	0,5–1,0	1,0–2,0	2,0–5,0	5,0–10	>10
	Drug room, floor, below bench beside compounding bench, before cleaning		X					
	Drug room, drug shelf (Trimetoprim)		X					
	Drug room, floor, below compounding bench, after cleaning		X					
Diaveridine	Drug room, bench beside compounding bench, before cleaning		X					
	Drug room, sink, before cleaning		X					
	Drug room, floor, below bench beside compounding bench, before cleaning		X					
	Drug room, drug shelf (Trimetoprim)		X					
	Drug room, floor, below compounding bench, after cleaning		X					
	Drug room, floor, below bench beside compounding bench, after cleaning		X					
	Patient toilet, floor, below toilet, after cleaning		X					
	Drug room, compounding bench, after cleaning with M-Ytdes45+		X					
	Drug room, compounding bench, after cleaning with water		X					
Doxycycline	Drug room, bench beside compounding bench, before cleaning		X					
	Drug room, sink, before cleaning		X					
	Drug room, floor, below bench beside compounding bench, before cleaning		X					
	Drug room, floor, below compounding bench, before cleaning		X					
	Drug room, floor, below bench beside compounding bench, after cleaning	X						

TABLE 6—*Continued*

Substance	Place	Level, ng/cm^2						
		0,01–0,1	0,1–0,5	0,5–1,0	1,0–2,0	2,0–5,0	5,0–10	>10
	Drug room, floor, below compounding bench, after cleaning	X						
Fluconazol	Drug room, floor, below bench beside compounding bench, before cleaning	X						
	Drug room, floor, below compounding bench, after cleaning	X						
	Patient toilet, floor, below toilet, before cleaning	X						
	Drug room, compounding bench, after cleaning with M-Ytdes45+	X						
	Drug room, compounding bench, after cleaning with water	X						
	Drug room, bench beside compounding bench, after cleaning with M-Ytdes45+	X						
Metronidazol	Drug room, floor, below bench beside compounding bench, before cleaning	X						
	Patient toilet, floor, below toilet, before cleaning		X					
	Drug room, waste container	X						
	Drug room, compounding bench, after cleaning with M-Ytdes45+		X					
	Drug room, compounding bench, after cleaning with water		X					
Ofloxacin	Drug room, bench beside compounding bench, before cleaning	X						
	Drug room, sink, before cleaning	X						
	Drug room, floor, below compounding bench, before cleaning	X						
	Drug room, floor, below compounding bench, after cleaning	X						
Trimethoprim	Drug room, compounding bench, before cleaning	X						

TABLE 6—*Continued*

Substance	Place	Level, ng/cm^2						
		0,01–0,1	0,1–0,5	0,5–1,0	1,0–2,0	2,0–5,0	5,0–10	>10
	Drug room, bench beside compounding bench, before cleaning	X						
	Drug room, sink, before cleaning	X						
	Drug room, floor, below compounding bench, before cleaning	X						
	Drug room, floor, below bench beside compounding bench, before cleaning	X						
	Drug room, drug shelf (Trimetoprim)	X						
	Drug room, floor, below compounding bench, after cleaning	X						
	Drug room, bench beside compounding bench, after cleaning		X					
	Patient toilet, floor, below toilet, before cleaning	X						
	Patient toilet, floor, below toilet, after cleaning	X						
	Drug room, compounding bench, after cleaning with M-Ytdes45+	X						
	Drug room, compounding bench, after cleaning with water	X						

The suggested and implemented measures have given some effect in reducing the spill and leakage as well as the distribution of emerging spill and leakage. Rather simple measures can contribute to decrease the spill and leakage that occur during compounding and other handling of antibiotic drugs. The first screening showed that it is possible to make compounding with almost no spill and leakage, i.e., by employing closed systems. One important outcome of this study has been an emerging awareness of the problem with spill and leakage and an ongoing discussion at the wards on how to improve the compounding procedures to reduce the drug spill and leakage.

During the visits at the wards during the follow-up screening, the staff showed an awareness that spill and leakage could occur and cause problems. This awareness contributed to a more concerned attitude, which influenced the individuals' method of working in a positive direction.

Cleaning Efficiency

An investigation of the cleaning efficiency was also carried out during the follow-up screening. Wipe samples were collected from various surfaces immediately before and after normal cleaning in drug rooms and patient toilets. All three wards had, according to the suggested measures, implemented cleaning of the benches with cleaning alcohol solution (M-Ytdes45+) and compounding on bench covers with plastic bottom. They also used a filter-spike for venting drug vials during compounding. None of the wards had, however, implemented a closed system for compounding. In all cases, the floors in the drug rooms and patient toilets were cleaned once every weekday with a humidified mop, where the mop cloth was changed for each room. The used mop cloths were washed in a laundry and reused until worn out. The recommended double mopping had not been implemented at any ward. Nor had an increased cleaning frequency been implemented. According to the current handbook for hospital care [21], all medical staffs are expected to clean visible spill and leakage in between regular cleaning occasions. The result from this investigation is presented in Table 7.

At Hospital 1, Surgery ward, a comparison of the contamination on the floor in the drug room before and after cleaning was carried out. Five different substances were found in the samples. The result shows that the same amounts of the substances were found after as found before the cleaning.

The comparison at Hospital 14, Hematology ward, comprised floors in the drug room and a patient toilet as well as two benches in the drug room used for compounding. On the floor in the drug room, four substances were found and two or three substances on other surfaces. The result shows that there are substances remaining on the surfaces after cleaning. On the benches about 50–70% of the original concentration of the substances was left after cleaning. On the floor in the patient toilet, it was the same level of substances before and after cleaning and about 50% of the original concentration of the substances was left on the floor in the drug room after cleaning.

At Hospital 15, General ward, wipe samples were collected at two places on the floor in the drug room and on the floor in one patient toilet. Wipe samples were also collected before and after cleaning on benched for compounding both

TABLE 7—*Result from the cleaning efficiency investigation.*

	Hospital 1, Surgery ward		Hospital 14, Hematology ward								Hospital 15, General ward								
	Drug room, floor		Drug room, floor		Left compounding bench		Right compounding bench		Patient toilet, floor		Drug room, floor first pos		Drug room, floor second pos		Patient toilet, floor		Compounding bench		
Antibiotic substance	Before	After M	Before	After M	Before	After M	Before	After M	Before	After M	Before	After M	Before	After M	Before	After M	Before	After w	After M
Cefalexin	ND	ND	0.03	0.03	0.03	0.026	0.03	0.03	0.03	0.04	ND	0.14	0.09	0.04	ND	0.01	ND	0.02	ND
Ciprofloxacin	0.01	0.01	ND	ND	ND	ND	ND	ND	ND	ND	0.04	ND	0.03	0.02	0.11	ND	ND	0.19	0.10
Diaveridin	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	0.01	0.05	0.08	0.02	0.01	0.02	0.02	0.01	0.01
Enrofloxacin	0.02	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Flukonazol	0.06	0.06	0.16	0.06	ND	ND	0.05	0.01	0.24	0.22	ND	ND	ND	ND	ND	ND	ND	ND	ND
Metronidazol	0.77	0.82	0.05	0.01	ND	ND	ND	ND	ND	ND	ND	ND	0.01	ND	0.19	ND	ND	0.12	0.20
Norfloxacin	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	0.03	0.02	0.02	0.01	ND	ND	ND	ND	ND
Trimetoprim	0.07	0.10	0.10	0.05	0.02	0.01	0.07	0.05	0.21	0.32	0.06	0.08	0.08	0.11	0.01	0.02	0.08	0.04	0.02

Note: Substance trivial names according to FASS [15]. Samples collected before and after cleaning with W–water or M–M-Ytdes45+, ND–not detected. All values in ng/cm^2.

on a surface that was clean with water, on a surface cleaned with cleaning alcohol solution (M-Ytdes45+). After the floor cleaning, some substances were absent, e.g., Ciprofloxacin, while others were found, e.g., Cefalexin. The levels of the substances decreased slightly for some substances, e.g., Diaveridin and Metronidazol. At the same time, the level also increased after the floor cleaning for other substances, e.g., Trimetoprim. The cleaning of the benches gave a more uncertain result. Only two substances were found before cleaning, while after water cleaning, five substances were found, and after cleaning with a cleaning alcohol solution (M-Ytdes45+), five substances were found.

At this investigation, wipe samples were collected at two adjacent surfaces before and after cleaning. If there had been small very local spill it could have been present on only one of the surfaces. This may explain why, occasionally, a substance could be found in one sample but not the other.

This investigation shows that the cleaning methods used need to be improved to efficiently remove spill and leakage of antibiotics. Cleaning bench surfaces with water or cleaning alcohol solution (45%) with a tenside gave similar result but did not manage to completely remove spill and leakage. Wiping the surfaces two times and changing the wipe tissue in between might improve the result.

The floor cleaning gave poor result why it is particularly important to improve the floor cleaning methods. Doubled mopping of the floor with change of mop cloth may improve the cleaning result. Doubled mopping was, however, not used at these occasions, since none of the wards had implemented that as normal cleaning procedure. Today a humidified mop is used. To improve the solubility of the drugs, it might be necessary to use more water when cleaning floors where drug contamination can occur.

These simple measures may improve the cleaning result. The cleaning methods at Swedish hospitals are similar, and it could be advisable to carry out a coordinated study to develop suitable cleaning methods to efficiently remove spill and leakage of antibiotics.

Occupational Hygiene Aspects

The result from this study shows that spill and leakage of antibiotics normally occur in not insignificant amounts. The screening, however, also showed that it is possible to handle antibiotics in medical care with only very small spill and leakage, particularly when closed systems were employed (e.g., PhaSeal or Tevadaptor).

The Swedish Work Health Authority Ordinance on Handling Cytostatics and other drugs with persistent toxic effects [5] also cover several antibiotic drugs, e.g., Penicillins, Cephalosporins, and some β-lactams. This ordinance comprise regulations on how these drugs should be handled during compounding and administration. It also covers waste handling and requirements for proper training and technical facilities.

Cefadroxil and Cefalexin are cephalosporins that were analyzed in this study. The result also shows that spill and leakage of these substances occur.

According to the Ordinance [5], these drugs should be handled in such way that the staff do not become occupational exposed. Moreover, the technical

systems used for compounding and administration should be tested for leakage in every day work on a regular basis. Also, waste handling shall be organized in such way that spill and leakage is minimized so no exposure occur. The staff shall also get adequate education to be able to handle the drugs in a safe way.

Consequently, it is, from occupational hygiene viewpoint, important to more systematically control spill and leakage of antibiotics in every day work. If such controls show that spill and leakage occur and that the staff becomes exposed, implementation of closed handling systems ought to be considered, especially for those drug types listed in the Ordinance [5].

Conclusions

The method used in this study is efficient and rapidly gives an extended picture of the spill and leakage that occur. With simultaneous determination of twelve different antibiotic substances that are active component in frequently used antibiotics today, the method gives a good view over the distribution of spill and leakage in most situations where antibiotics are handled.

The screening showed that spill and leakage of antibiotics occur. At least one substance in at least one sample was found at all wards that participated in the screening. The substances that were found in highest levels, all were active substances in drugs frequently administered both as tablets and as infusion. However, substances that were active substance in drugs only administered as tablets were also found. This means that also handling tablets must be considered when assessing spill and leakage of antibiotics as well as the staff exposure risks.

The wards that participated in the screening were classified into four groups depending on the contamination level. In this classification the following parameters were considered (i) the number of substances found; (ii) the number of samples with substances; and (iii) the level of contamination. Three of the 21 wards were classified as *Very high*, eight as *High*, six as *Medium,* and four as *Low*.

Different compounding systems affect the proportions of the spill and leakage. Employing a closed system will efficiently reduce the spill and leakage, while the use of a venting system without any filter gives large spill and leakage. The three wards that regularly used closed systems were all among the five wards with the lowest contamination level. In the same time, all three wards that used open venting systems, all were among the five wards with the highest contamination level.

Simple measures, such as using disposal gloves and bench covers and change these between each compounding and cleaning benches and other surfaces with cleaning alcohol (45 %) with tenside instead of 70% alcohol, all contribute to reduce the spill and leakage that occur as well as the distribution of the emerging spill and leakage. At all wards that participated in the follow-up screening, the contamination levels with antibiotics had decreased after implementation of preventive measures. The first screening, however, showed that it is possible to obtain almost insignificant contamination levels. It is,

consequently, important to discuss how to find adequate and safe procedures for handling antibiotics to minimize the spill and leakage, e.g., by employing closed systems.

Measurements before and after cleaning showed that only minor part of the contamination were removed with the regular cleaning procedures used today. It is therefore important to improve the cleaning methods. The cleaning methods at Swedish hospitals are similar. It can, thus, be appropriate to carry out a coordinated investigation to improve the cleaning methods to remove drug contamination on various surfaces.

Several of the substances, found in the screening, are covered by the Swedish Work Health Authority Ordinance [5]. According to the Ordinance handling of these drugs should be in such way that the staff does not become exposed to these drugs. Moreover, the handling systems used shall be controlled for spill and leakage in every day work. From occupational hygiene point of view, a more systematic control of spill and leakage is recommended. If such controls show that spill and leakage occur, implementation of closed system ought to be considered.

Acknowledgments

The writers acknowledge Arbetslivets Försäkringsaktiebolag (AFA) *[Worklife Insurance Inc.]* for the financial support that made this study possible.

APPENDIX A: INTRODUCTORY SURVEY FOR THE PROJECT

(**Translated from Swedish)**

Investigation of spill, leakage and staff exposure during handling of anitbiotics in hospital care

Running no X

1. We will participate in the project (circle appropriate)Yes, will participate No, will not participate
2. Unit (Hospital and ward)
   _____
3. Contact person (name, telephone, e-mail)
   _____
4. No of beds at the unit
   _____
5. No of staff that handles antibiotics
   _____
6. No of staff that nurse antibiotic treated patients
   _____
7. Total no of staff at the unit
   _____

8. How are antibiotics handled? (Circle correct (Yes or No) for each alternative)
   (a) We compound all antibiotics for infusion at the ward Yes No
   (b) We get all antibiotics from central compounding (e.g., pharmacy) Yes No
   (c) We both compound ourselves and get from central compounding Yes No
   (d) We compound antibiotics for bolus injections Yes No
   (e) We get prepared syringes for bolus injection from central compounding Yes No
   (f) We split tablets Yes No

9. List the five most frequently handled antibiotics and estimate the volumes (e.g., no of doses/week). Please also state the administration routes for each drug

_____

Please fill in this form and return it in the added prepaid envelope.
Thank you for your participation

APPENDIX B: DESCRIPTION OF ALL SAMPLING LOCATIONS AND ALL WIPED SURFACES AT EACH WARD

Biological Safety Cabinet (BSC); Frame—a home made plastic frame encompassing 100 cm^2 (for details see section "Material and methods")

Hospital no/Ward	Sample location	Wiped surface
1/Intensive care	Drug room, inner bench, left side	frame 10 × 10 cm
	Drug room, inner bench, middle	frame 10 × 10 cm
	Drug room, outer bench, left side	frame 10 × 10 cm
	Drug room, outer bench, right side	frame 10 × 10 cm
	Drug room, floor below outer bench	frame 10 × 10 cm
	Drug room, drug shelf above outer bench	frame 10 × 10 cm
	Cleaning room, sink at "dirty" side of bowl	frame 10 × 10 cm
	Cleaning room, floor below drug waste container	frame 10 × 10 cm
	Cleaning room, drug waste container	Lid and handle 30 × 40 cm
	Surveillance room, floor by door to drug room	frame 10 × 10 cm
1/Surgery	Drug room, sink for preparation, side of bowl	frame 10 × 10 cm
	Drug room, sink for preparation, bottom of bowl	Whole bottom 20 × 40 cm

Hospital no/Ward	Sample location	Wiped surface
	Drug room, work bench below drug shelf	frame 10 × 10 cm
	Drug room, floor below sink for preparation	frame 10 × 10 cm
	Cleaning room, sink for measuring urine volume at dirty side of bowl	frame 10 × 10 cm
	Cleaning room, drug waste container	Lid and handle 30 × 40 cm
	Cleaning room, floor below drug waste container	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, hand basin	Whole bowl 40 × 30 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
2/Hematology	Drug room, BSC inside middle	frame 10 × 10 cm
	Drug room, shelf for drugs	frame 10 × 10 cm
	Drug room, drug waste container	Lid and handle 30 × 40 cm
	Drug room, floor below BSC	frame 10 × 10 cm
	Patient ward, drug waste container	Lid and handle 30 × 40 cm
	Cleaning room, sink at “dirty” side of bowl	frame 10 × 10 cm
	Cleaning room, drug waste container	Lid and handle 30 × 40 cm
	Cleaning room, floor below drug waste container	frame 10 × 10 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 10 × 60 cm
2/Infection	Drug room, laminar flow bench 1, middle	frame 10 × 10 cm
	Drug room, laminar flow bench 2, middle	frame 10 × 10 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Drug room, needle waste box	Lid10 × 10 cm
	Drug room, drug waste container	Lid and lid handle 30 × 40 cm
	Drug room, floor below laminar flow benches	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
	Cleaning room, sink front of needle waste box	frame 10 × 10 cm
	Cleaning room, floor below sink with needle waste box	frame 10 × 10 cm

Hospital no/Ward	Sample location	Wiped surface
3/Intensive care	Drug room, preparation bench	frame 10 × 10 cm
	Drug room, sink side of bowl	frame 10 × 10 cm
	Drug room, floor below preparation bench	frame 10 × 10 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Drug room, drug waste container	Top brim including folded bag 110 × 4 cm
	Cleaning room, sink opposite to drug waste container, side of bowl	frame 10 × 10 cm
	Cleaning room, drug waste containers	Top brim of two containers including folded bag 140 × 5 cm + 140 × 3 cm
	Cleaning room, floor below drug waste containers	frame 10 × 10 cm
	Cleaning room, shelf beside drug waste containers	frame 10 × 10 cm
	Coffee room/office, floor	frame 10 × 10 cm
4/Surgery	Drug room, preparation bench, to right	frame 10 × 10 cm
	Drug room, preparation bench to left	frame 10 × 10 cm
	Drug room, floor below preparation benches	frame 10 × 10 cm
	Drug room, sink side of bowl	frame 10 × 10 cm
	Drug room, drug waste containers	Top brim of two containers total 240 × 2 cm
	Drug room, floor below drug waste containers	frame 10 × 10 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
	Nurse office, floor below door to drug room	frame 10 × 10 cm
5/Hematology	Drug room, BSC inside middle	frame 10 × 10 cm
	Drug room, bench beside BSC	frame 10 × 10 cm
	Drug room, floor below BSC	Frame 10 × 10 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Room outside of drug room where waste is handled, top brim of Packtosafe waste sealing system	frame 10 × 10 cm
	Room outside of drug room where waste is handled, floor below Pactosafe	frame 10 × 10 cm

Hospital no/Ward	Sample location	Wiped surface
	Air lock to room for cytostatics, blue basket	frame 10 × 10 cm
	Room for cytostatics, BSC inside middle	frame 10 × 10 cm
	Room for cytostatics, top brim of Pactosafe	frame 10 × 10 cm
	Room for cytostatics, floor below BSC	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	Frame 10 × 10 cm
	Nurse office, floor below hand basin	frame 10 × 10 cm
6/Hematology-Oncology	Drug room, preparation bench left side	frame 10 × 10 cm
	Drug room, preparation bench right side	frame 10 × 10 cm
	Drug room, floor below preparation bench	frame 10 × 10 cm
	Drug room, drug waste container	Lid and lid handle 30 × 40 cm
	Drug room, drug waste container	Top of front side 30 × 20 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
	Nurse office, floor middle	frame 10 × 10 cm
7/Infection	Drug room, BSC inside middle	frame 10 × 10 cm
	Drug room, bench beside BSC	frame 10 × 10 cm
	Drug room, floor below BSC	frame 10 × 10 cm
	Drug room, floor below bench beside BSC	frame 10 × 10 cm
	Drug room, top drug waste container under BSC	Front side 30 × 30–25 cm (conical)
	Drug room, bottom drug waste container under BSC	Lid and lid handle 30 × 40 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
7/Hematology	Drug room, right preparation bench	frame 10 × 10 cm
	Drug room, left preparation bench	frame 10 × 10 cm
	Drug room, floor below preparation benches	frame 10 × 10 cm

Hospital no/Ward	Sample location	Wiped surface
	Drug room, drug waste container	Lid and lid handle 30 × 40 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Patient day room, floor	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
8/General ward	Drug room, preparation bench mid-room	frame 10 × 10 cm
	Drug room, preparation bench right	frame 10 × 10 cm
	Drug room, sink side of bowl	frame 10 × 10 cm
	Drug room, floor below preparation benches	frame 10 × 10 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Drug room, drug waste container	Lid and lid handle 30 × 40 cm
	Drug room, floor below drug waste container	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	frame10 × 10 cm
	Nurse office, floor below door to drug room	frame 10 × 10 cm
9/Infection	Drug room, preparation bench mid-room	frame 10 × 10 cm
	Drug room, preparation bench right	frame 10 × 10 cm
	Drug room, preparation bench left	frame 10 × 10 cm
	Drug room, floor below preparation bench mid-room	frame 10 × 10 cm
	Drug room, floor below drug waste container	frame 10 × 10 cm
	Drug room, drug waste container	Top brim and front side 1 × 140 cm + 30 × 60 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Air lock patient ward, drug waste container	Front of plastic bag 20 × 30 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
9/Hematology	Drug room, preparation bench middle	frame 10 × 10 cm
	Drug room, floor below preparation bench	frame 10 × 10 cm

Hospital no/Ward	Sample location	Wiped surface
	Drug room, floor below drug waste container	frame 10 × 10 cm
	Drug room, drug waste container	Lid and lid handle 30 × 40 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Drug room, small bench	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
10/Infection	Drug room, bench opposite to preparation bench	frame 10 × 10 cm
	Drug room, preparation bench	frame 10 × 10 cm
	Drug room, sink side of bowl	frame 10 × 10 cm
	Drug room, floor below preparation bench	frame 10 × 10 cm
	Drug room, drug waste container	Top brim and front side including folded plastic bag 1 × 160 + 30 × 20 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Drug room, floor below work bench opposite to preparation bench	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
11/Infection	Drug room, preparation bench to left	frame 10 × 10 cm
	Drug room, preparation bench to right	frame 10 × 10 cm
	Drug room, sink side of bowl	frame 10 × 10 cm
	Drug room, floor middle	frame 10 × 10 cm
	Drug room, waste sack holder	Lid on both sides 2 × 20 × 30 cam + edge 1 × 20 × 30 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Drug room, shelf to left of drug shelf	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
12/Surgery	Drug room, preparation bench by window middle	frame 10 × 10 cm
	Drug room, preparation bench opposite to window middle	frame 10 × 10 cm
	Drug room, preparation bench in the middle	frame 10 × 10 cm

Hospital no/Ward	Sample location	Wiped surface
	Drug room, floor in the middle	frame 10 × 10 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Drug room, drug waste container	Lid 31 × 24 cm
	Cleaning room, drug waste container	Lid and top brim 30 × 40 cm
	Cleaning room, floor below drug waste container	frame 10 × 10 cm
	Room for cytostatics, shelf with cytostatics	frame 10 × 10 cm
	Room for cytostatics, drug waste container	Top brim 1 × (30 + 30 + 40 + 40) cm
13/General ward	Drug room, preparation bench to left	frame 10 × 10 cm
	Drug room, preparation bench to right	frame 10 × 10 cm
	Drug room, floor below preparation benches	frame 10 × 10 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Drug room, trolley	frame 10 × 10 cm
	Cleaning room, drug waste container	Lid and top brim 40 × 40 cm
	Cleaning room, floor below drug waste container	frame 10 × 10 cm
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
14/Hematology	Drug room, preparation bench to left	frame 10 × 10 cm
	Drug room, preparation bench to right	frame 10 × 10 cm
	Drug room, floor below preparation benches	frame 10 × 10 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Waste room, drug waste container	Lid and lid handle 40 × 40 cm
	Waste room, cytostatics waste container	Lid and lid handle 40 × 40 cm
	Room for cytostatics, preparation bench	frame 10 × 10 cm
	Room for cytostatics, floor below preparation bench	frame 10 × 10 cm
	Room for cytostatics, floor below door	frame 10 × 10 cm

Hospital no/Ward	Sample location	Wiped surface
	Patient WC, toilet	Seat ring 7 × 60 cm
	Patient WC, floor below toilet	frame 10 × 10 cm
14/Infection	Drug room, preparation bench on left side	frame 10 × 10 cm
	Drug room, preparation bench on right side	frame 10 × 10 cm
	Drug room, floor below preparation benches	frame 10 × 10 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Drug room, drug waste container	Top brim $1 \times (26 + 26 + 36 + 36)$ cm
	Drug room, waste container in plastics	Lid and lid handle 40 × 40 cm
	Cleaning room, drug waste container	Top brim $1 \times (40 + 40 + 40 + 40)$ cm
	Cleaning room, floor below drug waste container	frame 10 × 10 cm
	Cleaning room, bench beside drug waste container	frame 10 × 10 cm
15/General ward	Drug room, preparation bench middle	frame 10 × 10 cm
	Drug room, bench beside preparation bench	frame 10 × 10 cm
	Drug room, sink beside bowl	frame 10 × 10 cm
	Drug room, floor below preparation bench	frame 10 × 10 cm
	Drug room, drug shelf	frame 10 × 10 cm
	Drug room, floor below sink	frame 10 × 10 cm
	Drug room, drug waste containers	Top brim of two containers $1 \times (23 + 23 + 19 + 19) + 1 \times (39 + 39 + 27 + 27)$ cm
	Cleaning room, drug waste container	Lid and lid handle 40 × 40 cm
	Cleaning room, floor below drug waste containers	frame 10 × 10 cm
16/General ward	Drug room, waste sack holder	Lid 31 × 24 cm
	Drug room, BSC inside middle	frame 10 × 10 cm
	Drug room, bench opposite to BSC	frame 10 × 10 cm
	Drug room, bench beside the door	frame 10 × 10 cm
	Drug room, floor below BSC	frame 10 × 10 cm
	Drug room, floor below bench by the door	frame 10 × 10 cm

Hospital no/Ward	Sample location	Wiped surface
	Drug room, drug shelf	frame 10 × 10 cm
	Drug room, drug waste container s	Top brim of two containers 2 × 1 × (39 + 39 + 26 + 26) cm
	Cleaning room, drug waste container	Top brim 1 × (40 × 40) cm
	Cleaning room, floor below drug waste container	frame 10 × 10 cm
	Room for cytostatics, BSC inside middle	frame 10 × 10 cm
	Room for cytostatics, floor below BSC	frame 10 × 10 cm
	Room for cytostatics, drug waste container	Lid and lid handle 40 × 40 cm

APPENDIX C: EXAMPLES OF PREVENTIVE MEASURES TO MINIMIZE THE OCCURRENCE OF SPILL AND LEAKAGE

- Consider to change to a closer compounding system. Several studies on spill and leakage during compounding cytostatics have shown that a closed system for compounding minimizes the drug spill and leakage. Also spikes with filter reduce spill and leakage compared with open systems [1].
- Hold the orifice of the tubing over a collection vessel or a bench cover sheet with plastic bottom when filling tubings in infusions systems and not over the floor, bench surface or a sink. Possible drug leakage through the orifice will be then collected on disposal material that can be discarded in a proper waste container, without any emerging spill onto surfaces.
- Prepare as much as possible in the drug room to minimize the risk for spill and leakage in the nursing rooms.
- Use a mixing device for sealed drug vials.
- When possible during administering of antibiotics in pre-compounded infusion bags, start by fill the tubing's of infusion systems with saline solution instead of drug solution before connection to patient infusion port and then end the administration with saline solution to empty all tubing's from the drug solution.

EXAMPLES OF MEASURES TO PREVENT SPATIAL DISTRIBUTION OF EMERGED SPILL AND LEAKAGE

- Use disposal gloves during compounding and change gloves after each compounding to avoid distributing possible spill from one infusion bag to next.

- Carry out the compounding on a bench cover sheet with plastic bottom, e.g., an examination sheet and change/discard the sheet between each compounding. Any spill on the sheet will then not be distributed to next compounding or to bench surfaces.
- Handling tablets can also contribute to distribution of antibiotic substances. To minimize the distribution of tablet dust use disposal gloves and handle the tablets on a bench cover sheet with plastic bottom. Discard the gloves and sheet when the task is finished.
- Discard disposal gloves before leaving the drug room to avoid distribution of drugs through contaminated gloves.
- In case of visible spill, always wipe up the spill everywhere it occur using disposal wipe material. Clean afterwards with water or cleaning alcohol (45% with detergent) before any disinfection with 70% alcohol.
- Most drugs dissolve better in water than 70% disinfection alcohol. Cleaning will, thus, be more efficient if all surfaces are cleaned with water or cleaning alcohol before any disinfection with 70% alcohol.
- If cleaning alcohol (45% with detergent) is regularly used for cleaning a thin film of detergent will be formed and cleaning with pure water will be required on a regular basis.
- Improved cleaning can be accomplished by wet mopping twice and change mop in between in places where spill and leakage can occur, e.g., drug room, patient toilettes. A more frequent cleaning can also be required. It is also important to wipe up visible spill and leakage between cleaning occasions and clean with cleaning alcohol or water.

References

[1] National Institute for Occupational Safety and Health, 2010, "Occupational Exposure to Antineoplastic Agents," http://www.cdc.gov/niosh/topics/antineoplastic/default.html (Last accessed October 20, 2010).

[2] *CSHP Guidelines for the handling and disposal of hazardous pharmaceuticals (including anti-cancer drugs)*, Canadian Society of Hospital Pharmacists, Ottawa, 1993.

[3] American Society of Health-System Pharmacists, "ASHP Guidelines on Handling Hazardous Drugs," *Am. J. Health Syst. Pharm.*, Vol. 63, 2006, pp. 1172–1193.

[4] HSE. Information Sheet MISC 615, 2003, "Safe Handling of Cytotoxic Drugs," *Health and Safety Executive*, http://www.hse.gov.uk/pubns/misc615.pdf (Last accessed October 20, 2010).

[5] Work Health Authority, Ordinance AFS 2005:5, "Cytostatics and Other Drugs With Persistent Toxic Effects," *Swedish Work Health Authority*, Stockholm, Sweden, 2005.

[6] Apotekens Service AB, "Försäljningsstatistik [Sale Statistics (in Swedish)]," http://www.apotekensservice.se/Om_statistik/ (Last accessed October 29, 2010).

[7] Niessen, W. M. A., "Analysis of Antibiotics by Liquid Chromatography–-Masspectrometry," *J. Chromatogr. A*, Vol. 812, 1998 pp. 53–75.

[8] Stolker, A. A. M. and Brinkman, U. A. Th., "Analytical Strategies for Residue Analysis of Veterinary Drugs and Growth-Promoting Agents in Food-Producing Animals—A Review," *J. Chromatogr. A*, Vol. 1067, 2005 pp. 15–53.

[9] Hirsh, R., Ternes, T., Haberer, K., and Kratz, K.-L., "Occurrence of Antibiotics in the Aquatic Environment," *Sci. Total Environ.*, Vol. 225, 1999 pp. 109–118.

[10] Kolpin, D. W., Furlong, E. T., Meyer, M. T., Thurman, E. M., Zaugg, S. D., Barber , and L. B., Buxton, H. T., "Pharmaceuticals, Hormones and Other Organic Wastewater Contaminants in US Streams 1999–2000: A National Reconnaissance," *Environ. Sci. Technol.*, Vol. 36, 2002, pp. 1202–1211.

[11] Lindberg, R., Jarnheimer, P. -Å. J.J., Olsen, B., Johansson, M., and Tysklind, M., "Determination of Antibiotic Substances in Hospital Sewage Water Using Solid Phase Extraction and Liquid Chromatography/Mass Spectrometry and Group Analogue Internal Standards," *Chemosphere*, Vol. 57, 2004, pp. 1479–1488.

[12] Lindberg, R., 2006, "Determination of Antibiotivs in the Swedish Environment with Emphasis on Sewage Treatment Plants," *dissertation*, Umeå Univeristy [ISBN 91-7264-045-6].

[13] Tuerk, J., Reinders, M., Dreyer, D., Kiffmeyer, T. K., Schmidt, K. G., and Kuss, H.-M., "Analysis of Antibiotics in Urine and Wipe Samples From Environmental and Biological Monitoring—Comparison of HPLC With UV-, Single MS- and Tandem MS-detection," *J. Chromatogr. B*, Vol. 831, 2006, pp. 72–80.

[14] Nygren, O. and Lindahl, R., "Development and Validation of a Method for Screening Spill and Leakage of Antibiotics on Surfaces Based on Wipe Sampling and HPLC-MS/MS Analysis," *J. ASTM Int.*, (submitted).

[15] FASS, 2010, "Farmaceutiska Specialiteter i Sverige (Pharmaceutical Specialites in Sweden], (in Swe) Läkemedelsinsdustriföreningen, "http://www.fass.se (Last accessed October 25, 2010).

[16] Nygren, O. and Aspman, O., "Validation and Application of Wipe Sampling and Portable XRF Analysis as an on-site Screening Method for Assessment of Deposited Aerosols on Workplaces," *Aust. J. Chem.*, Vol. 37, 2004 pp. 1021–1028.

[17] Nygren, O., "Wipe Sampling as a Tool for Monitoring Aerosol Deposition in Workplaces," *J. Environ. Monit.*, Vol. 8, 2006, pp. 49–52.

[18] Hornung, R. W. and Reed, L. D., "Estimation of Average Concentration in the Presence of Nondetectable Values," Appl. Occup. Environ. Hyg., Vol. 5, 1990, pp. 46–51.

[19] Fransman, W., Vermeulen, R., and Kromhout, H., "Dermal Exposure to Cyclophosphamide in Hospitals During Preparation, Nursing and Cleaning Activities," *Int. Arch. Occup. Environ. Health*, Vol. 78, 2005, pp. 403–412.

[20] Touzin, K., Bussières, J. F., Langlois, É., Lefebvre, M., and Métra, A., "Pilot Study Comparing the Efficacy of Two Cleaning Techniques in Reducing Environmental Contamination With Cyclophosphamide," *Ann. Occup. Hyg.*, Vol. 54, 2010, pp. 351–359.

[21] SKL, "Handbok för hälso- och sjukvård. Städning. (Handbook for hospital care. Cleaning) (in Sweden) Sveriges Kommuner och Landsting," 2008-10-08, http://www.sjukvardsradgivningen.se/handboken (Last accessed October 27, 2010).

GENERAL TOPICS

Reprinted from JAI, Vol. 8, No. 9
doi:10.1520/JAI103523
Available online at www.astm.org/JAI

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Application of ASTM Standard Practice D6602 in the Investigation of Outdoor Environmental Surface Particulate Including Darkening Agents

ABSTRACT: ASTM D6602 "Standard Practice for Sampling and Testing of Possible Carbon Black Fugitive Emissions or Other Environmental Particulate, or Both" was developed for the investigation of outdoor sooty surface problems and distinguishing manufactured carbon black, in the N100 to N900 series, from other environmental particulates. It is a microscopy technique that is well suited for the study of darkening agent complaints. Samples are collected from surfaces with cotton balls, wipes, and tape-lifts. The particulate is then analyzed by light microscopy and electron microscopy. The practice is used as a means to differentiate between darkening caused by fungal growth (biofilms), soots, or other dark particles. Polarized light microscopy provides information about the relative amounts of the different types of particles present and some information about the type of biofilm, if present. Transmission electron microscopy provides confirmation of aciniform soot, if present, and some information about the possible source of the soot. An investigation of dark surface particulate in a city where both a carbon black manufacturing facility and an oil refinery were possible industrial sources provides an illustration of the use of the method.

KEYWORDS: soot, carbon black, microscopy, outdoor surface sampling

Manuscript received October 25, 2010; accepted for publication July 14, 2011; published online August 2011.

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Cite as: Millette, J. R., Rook, H. L. and Compton, S., "Application of ASTM Standard Practice D6602 in the Investigation of Outdoor Environmental Surface Particulate Including Darkening Agents," *J. ASTM Intl.*, Vol. 8, No. 9. doi:10.1520/JAI103523.

Introduction

To protect the public health and welfare, the United States Environmental Protection Agency has established national ambient air quality standards (NAAQS) under the Clean Air Act. In addition to the primary NAAQS which protect the public health, secondary NAAQS are established to protect the public from adverse effects such as materials damage and surface soiling by particulate matter (PM). Soiling caused by the deposition of PM can reduce the aesthetic appeal of buildings and culturally important articles such as statues and works of art. Particle-related soiling can result in increased cleaning frequency and repainting, and may reduce the useful life of the soiled materials [1]. In the USEPA NAAQS documents [2] "soiling" is described as the deposition of particles on surfaces by impingement and the accumulation of particles on the surface of an exposed material that results in degradation of its appearance.

Unfortunately, very little data are available in the published scientific literature concerning the composition, characteristics and quantities of particles that are involved in episodes of soiling of outdoor surfaces. The most common visual effect of soiling is a darkening of the surface. Common outdoor surface darkening agents include fungal material (biofilms), soot, plant fragments, rubber particles, and soil minerals. In some cases rust can also be a significant cause of darkening.

Two ASTM Standards, ASTM D3274 [3] and ASTM D4610 [4], that deal with discoloration of painted surfaces and the ASTM D6602 "Standard Practice for Sampling and Testing of Possible Carbon Black Fugitive Emissions or Other Environmental Particulate, or Both" [5] can be used for the investigation of particulate soiling or darkening on outdoor surfaces. The particulate is collected from the surface with cotton balls, wipes, and/or tape lifts. It is then analyzed by light microscopy and electron microscopy. Polarized light microscopy (PLM) provides information about the relative amounts of the different types of particles present and transmission electron microscopy (TEM) provides confirmation characterization of aciniform carbon, if present. Scanning electron microscopy (SEM) equipped with x-ray elemental analysis can help identify black, opaque particles such as coal and coke.

A study involving a large number of outdoor surface samples collected over 12 years provides a useful illustration of the use of the microscopy methods in ASTM D6602 in investigations of soiled or darkened surfaces.

Method

ASTM D6602 was developed for the investigation of outdoor sooty surface problems and distinguishing manufactured carbon black from soot and other environmental particulates. No other published standard for characterizing a variety of darkening agents in environmental samples is available. According to ASTM D6602, the use of a transmission electron microscope (TEM) is mandatory (Ref. [5], Sec. 7.1) in identifying the presence of carbon black in a sample. However, the mandatory section, by itself, does not provide a sufficient analysis to determine plausible causes of darkening from particles on a surface. To declare that the darkening on the surface at a particular location is caused by

carbon black, it is not sufficient to identify a few carbon black aggregates in the sample by TEM. If a surface is covered with a high amount of fungal growth (biofilm), dark soil minerals or soot that contains a trace of carbon black, the use of only the mandatory section would result in only a report of a positive for carbon black even though the actual cause of the darkening was obviously biofilm, dark soil minerals or soot when the overall sample was considered. To gain a full appreciation about the contributions of various sources to soiling or darkening requires the use of the non-mandatory light microscopy part of the procedure (Ref. [5], Sec. 8.3). Utilizing a polarized light microscope (PLM), the analyst is instructed to estimate and record the percentage of each type of component found (Ref. [5], Sec. 8.3.3.3).

The full D6602 analysis used to investigate the significant causes of darkening in a particular property involves three tasks:

1. Classification of the particles into categories by PLM.
2. A semi-quantitative determination of the relative amounts of the various components by PLM.
3. Distinguishing between manufactured carbon black and aciniform carbon particles by TEM.

The identification of environmental particles and classification into categories by PLM has been described previously [6–8]. The microscopist relies on his/her training, experience and reference to various published articles and books about particle characteristics [9–18]. Some representative PLM images of particles in the common darkening agent classes of soot, fungal growth (biofilms), soil minerals, plant fragments, and rubber particles are shown in Figs. 1–6.

The semi-quantitative visual estimate of percentages of particles in a sample is a well known technique that has been used for many years by geologists [20–23], paleontologists [24], and asbestos analysts [25,26]. Analysts learn to perform calibrated visual estimates by studying comparison charts where a

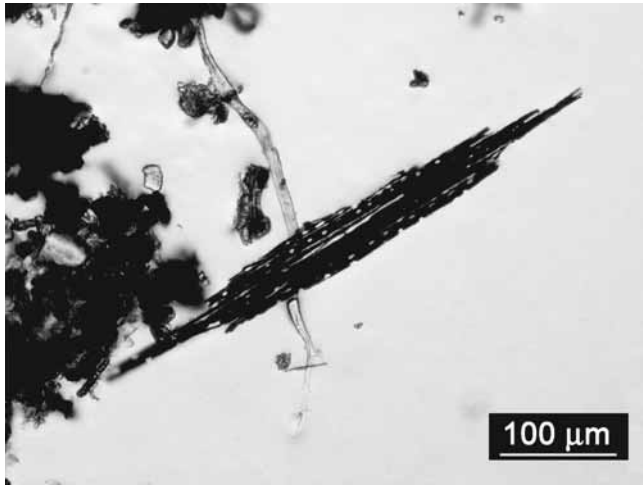

FIG. 1—*PLM image of soot (char form).*

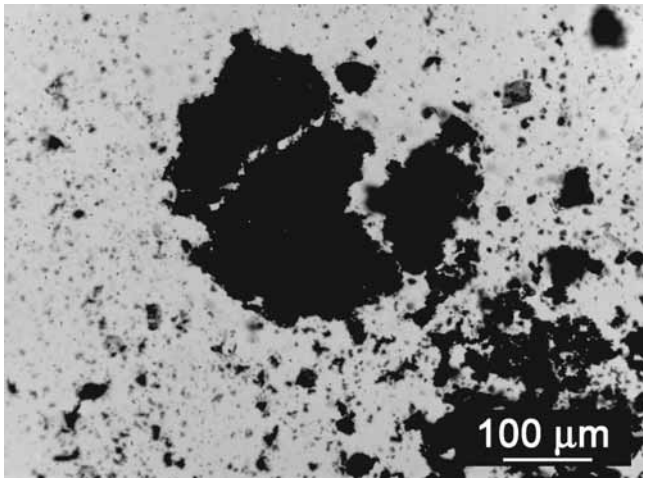

FIG. 2—*PLM image of soot (aciniform).*

known percentage of the particles in the chart has been filled in with dots or other dark figures. Figure 7 shows a representative comparison chart used for calibrating analysts in semi-quantitative visual estimation. Samples of known composition made from known volumes of various components are also used to "calibrate" an analyst. A relative standard error of 30 % was found in an inter-laboratory study of semi-quantitative visual estimates of surface darkening agents [27]. Although a 30 % error rate may seem high, it is quite good for semi-quantitative techniques. The finding of a 30 % error rate for surface dust

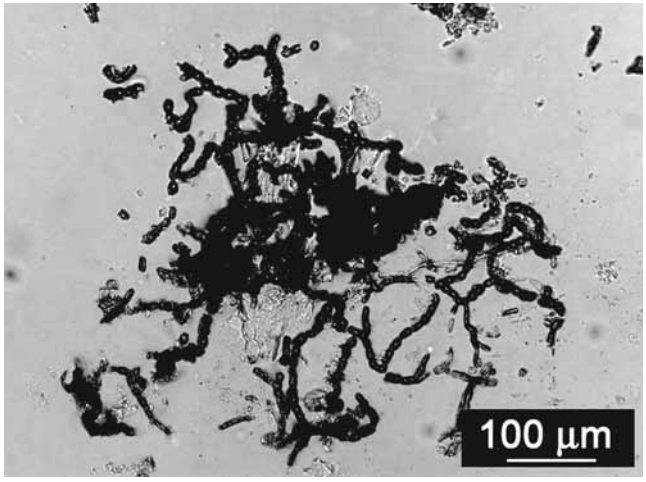

FIG. 3—*PLM image of fungal spores and hyphae.*

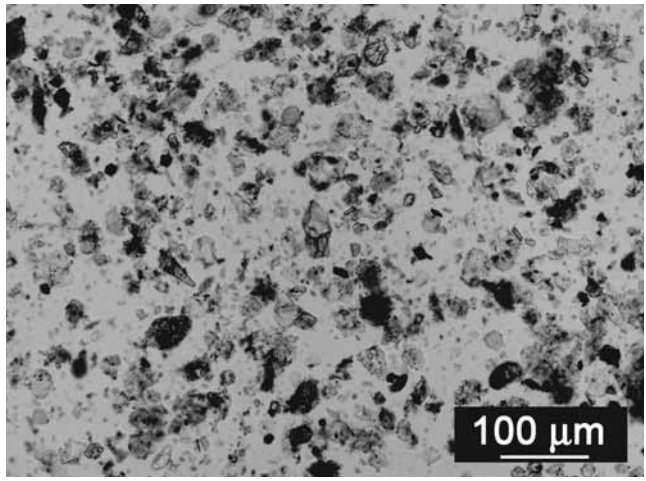

FIG. 4—*PLM image of soil minerals.*

darkening agents was consistent with the error rates for inter-laboratory tests of visual estimates used in asbestos analysis. The error rates for three standard methods of asbestos analysis where semi-quantitative visual estimates were used were reported to be 32.2, 25.7, and 26.9 % [28].

Distinguishing between carbon black and other aciniform carbon particles cannot be done at the light microscope level. It requires a TEM and consulting the mandatory section of ASTM D6602. The microscopist relies on his/her training, experience and reference to various published articles and books about

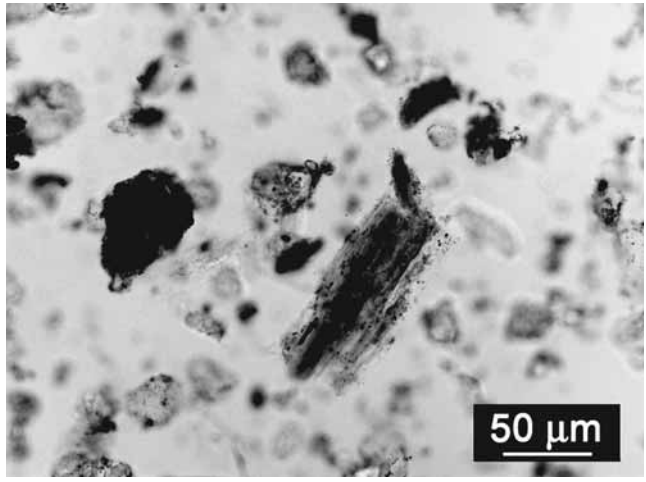

FIG. 5—*PLM image of plant fragments.*

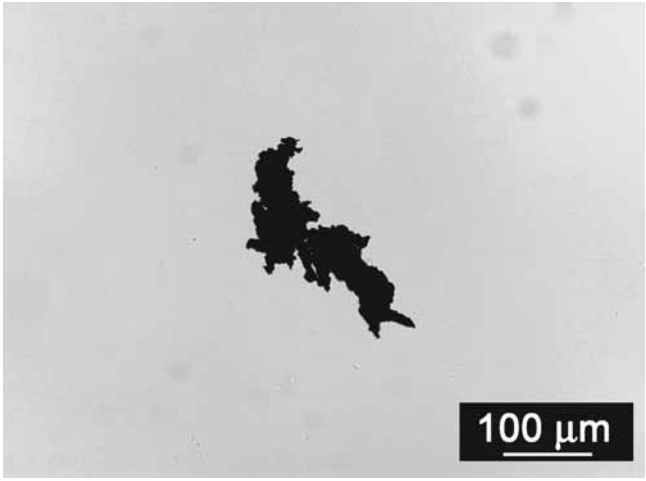

FIG. 6—*PLM image of rubber particles*.

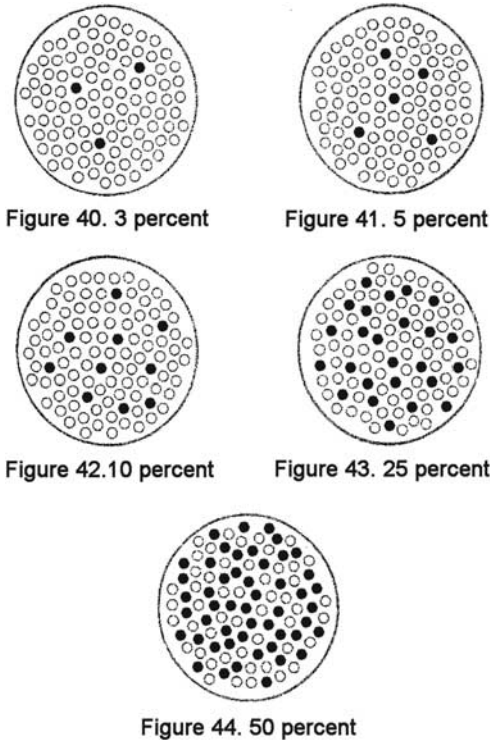

FIG. 7—*Representative comparison chart used to calibrate analysts in semi-quantitative visual estimates*.

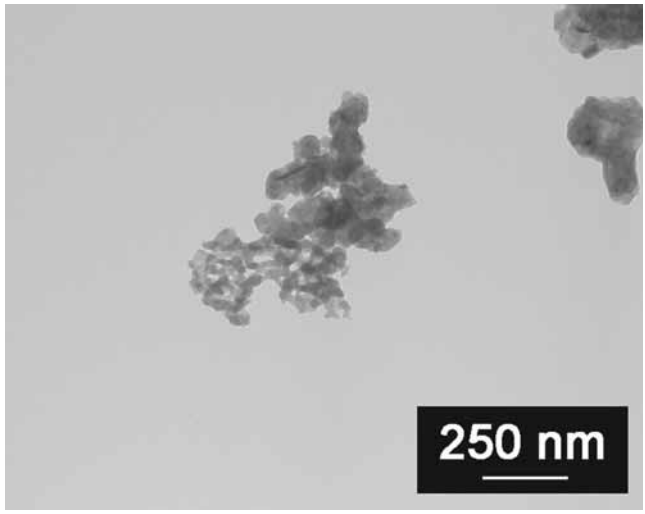

FIG. 8—*TEM image of aciniform soot from a vegetative fire.*

soot particle characteristics [29–36]. Known reference samples of carbon black [37] are available as well as a NIST standard reference sample of diesel soot [38]. Some representative TEM images, x-ray spectra and primary particle size distributions for aciniform carbon particles are shown in Figs. 8–16.

Some Results from a Study of Outdoor Surface Particulate

This study involved the analysis of 587 outdoor surface samples collected in a city in Oklahoma and surrounding areas over the period from 1998 to 2010. The

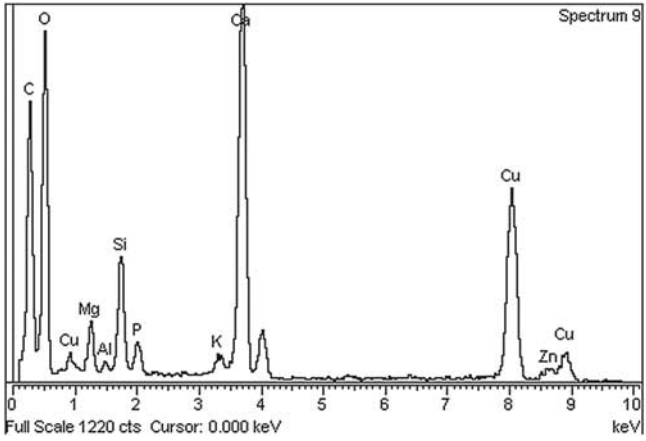

FIG. 9—*X-ray spectrum of aciniform soot from a vegetative fire.*

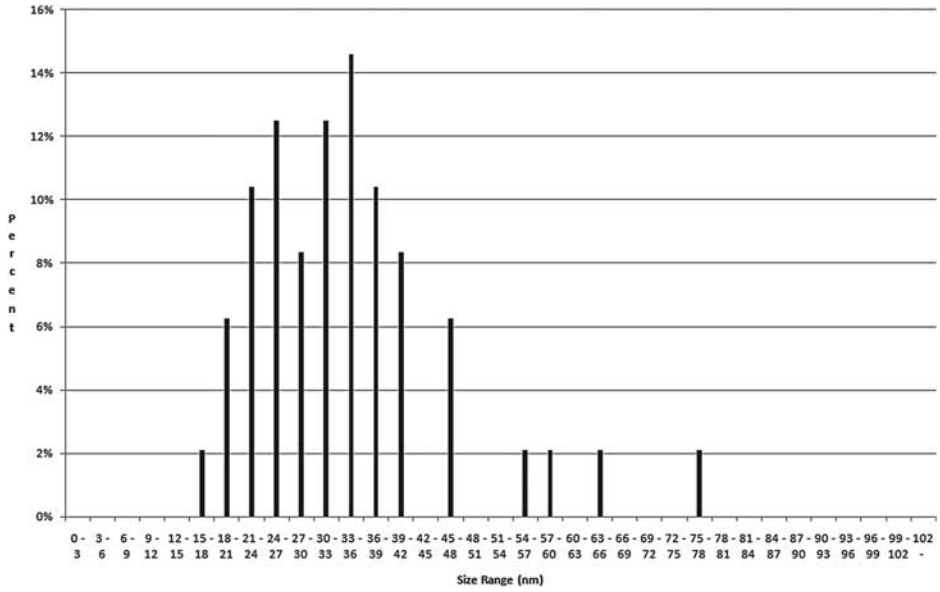

FIG. 10—*Primary particle size distribution of aciniform soot from a vegetative fire.*

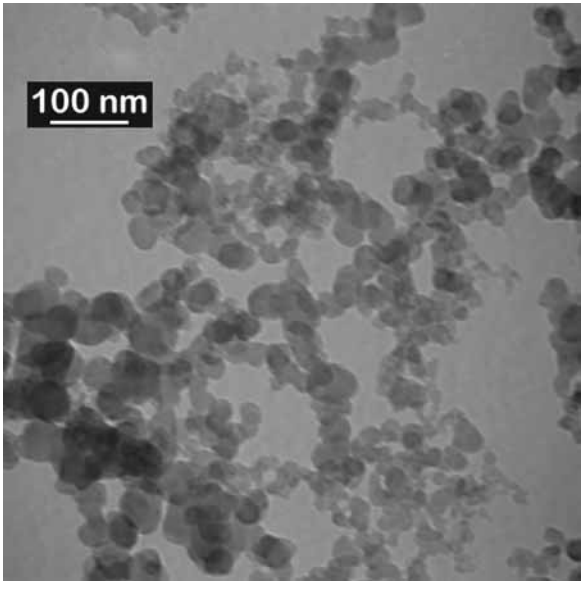

FIG. 11—*TEM image of aciniform soot from diesel exhaust (NIST SRM 2975).*

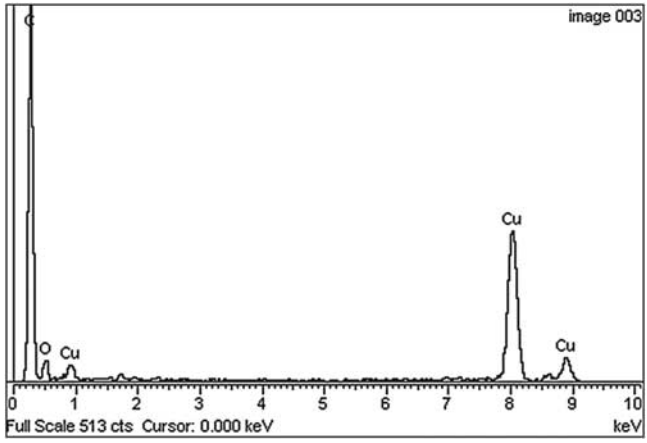

FIG. 12—*X-ray spectrum of aciniform soot from diesel exhaust (NIST SRM 2975).*

samples were analyzed following the ASTM D6602 Standard including the non-mandatory PLM and mandatory TEM sections. The study was conducted in conjunction with several lawsuits involving allegations that emissions from a carbon black plant caused surface darkening (soiling) at residential and commercial properties in the city area. All of the samples were complaint samples, surface samples from properties where there was a claim of darkening or surface particle soiling. This presentation is not intended to be a full case study of the surface darkening on properties in the city area. Such a study might include

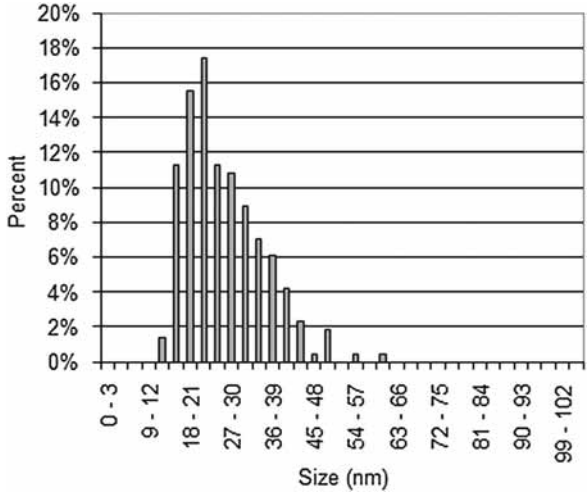

FIG. 13—*Primary particle size distribution of aciniform soot from diesel exhaust (NIST SRM 2975).*

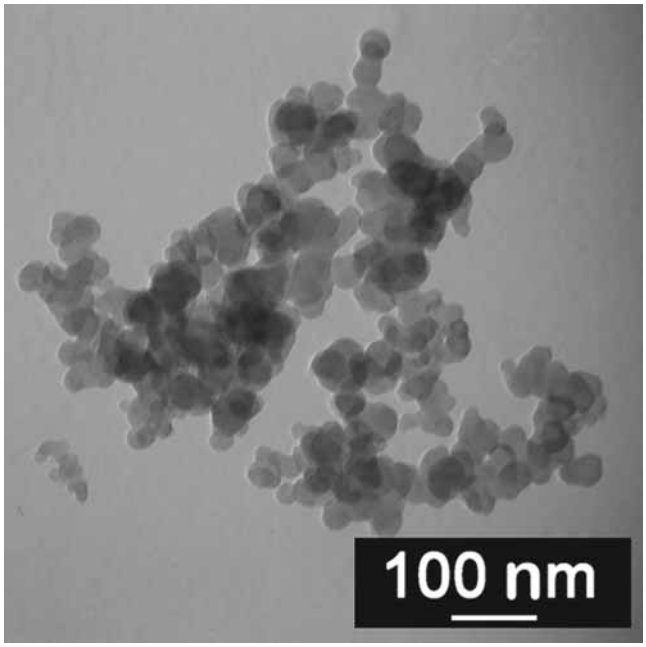

FIG. 14—*TEM image of aciniform soot from N326 carbon black (ASTM SRB 8A).*

air dispersion modeling and other factors. This presentation indicates the value of using the ASTM D6602 Standard used to analyze surface samples.

The city has a population of approximately 25,000. Industrial particulate material (PM) emission sources in the area include a petroleum refinery less

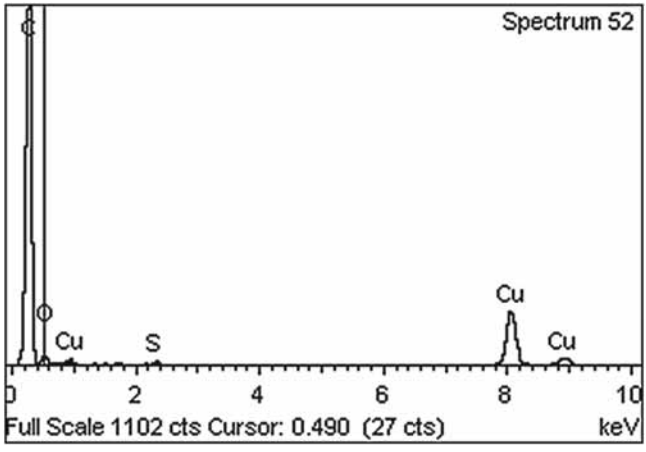

FIG. 15—*X-ray spectrum of aciniform soot from N326 carbon black (ASTM SRB 8A).*

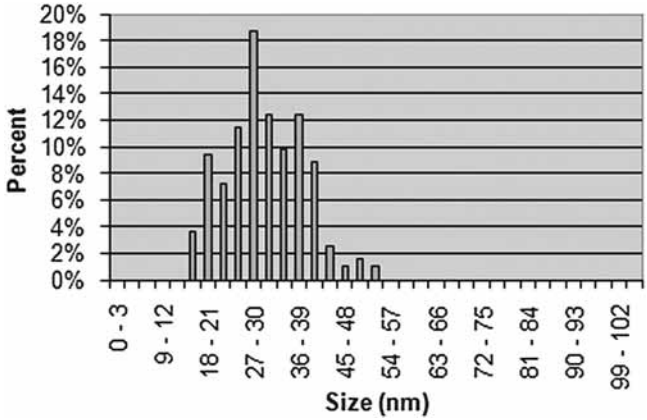

FIG. 16—*Primary particle size distribution of aciniform soot from N326 carbon black (ASTM SRB 8A).*

than 1 mile from downtown, a carbon black plant about 1.5 miles from the city, a coal-fired electric power generating station about 17 miles from the city and a plant that produces industrial grade calcined petroleum coke that is located about 40 miles from the city. Non-industrial sources of PM include roadway and railway traffic, trash burning on individual properties, and agricultural activities including ranching, soil tilling, crop cutting, and some periodic grass burning. Pollen granules, fungal spores, and decaying vegetation also contribute to the natural PM in the area.

The samples were collected using cotton balls, polyester wipes, and cellophane tape-lifts. The cotton balls were packaged in the laboratory in resealable plastic bags. The polyester wipes used in this study were 4 in. square clean-room wipes packaged at the laboratory in individual reclosable plastic bags. The 4 in. clean-room wipes come soaked in alcohol which prevents biological growth. They are dried prior to sample collection. The tape-lifts were prepared in the laboratory using clear cellophane tape attached to clean, glass microscope slides. The tape-lifts were also packaged in resealable plastic bags. The sampling team used particle-free disposable gloves when collecting the samples. The samples were collected from areas on the properties where the surface particulate appeared to be dark.

Table 1 shows examples of the results for several samples using the ASTM D6602 non-mandatory PLM. The values are percentage estimates by volume. The sample identification numbers have been coded. Table 2 shows a comparison of two samples that were split and sent to a second laboratory for the same analysis. The two laboratories were independent except for the fact that they both had extensive training in particle analysis by microscopy from the same McCrone Research Institute in Chicago, IL. Although there are differences in the trace components, the major component findings are similar.

Table 3 shows the average estimated percentage values on a year-by-year basis for the particle classes found in the surface samples during the study. Soil

TABLE 1—*Examples of results of PLM particle analyses. ND = not detected, trace = less than 1 % by volume.*

Particulate components	Sample 17ACHC	Sample 20AEHA	Sample 19AAIH	Sample 17AGACD	Sample 17BCBG
Pollen grains	ND	Trace–2 %	1–5 %	10–15 %	ND
Fungal, mold, biofilm	5–10 %	5–10 %	50–75 %	3–5 %	100 %
Soil minerals	15–30 %	30–50 %	15–25 %	60–70 %	ND
Soot	10–15 %	5–10 %	ND	2–3 %	ND
Flyash	ND	Trace	ND	ND	ND
Glass fibers	ND	ND	ND	Trace	ND
Plant fragments	20–30 %	ND	10–20 %	1–2 %	ND
Paint	ND	10–20 %	1–5 %	2–3 %	ND
Construction debris	ND	10–15 %	ND	ND	ND
Insect parts	ND	ND	Trace	Trace	ND
Starch	ND	Trace	ND	Trace	ND
Rust/metal flakes	15–20 %	1–3 %	ND	1–2 %	ND
Rubber	5–10 %	3–5 %	ND	2–3 %	ND
Petroleum coke/coal	5–10 %	ND	ND	Trace	ND
Catalyst	ND	Trace-2 %	ND	Trace	ND
Carbon black	~8 %	~3 %	ND	Trace	ND

TABLE 2—*Comparison of PLM particle classifications of 2 samples by two laboratories. ND = not detected.*

	Lab A	Lab B	Lab A	Lab B
Particulate components	Sample 22ACGA	Split of 22ACGA	Sample 22ACGB	Split of 22ACGB
Skin cells	ND	Trace	ND	ND
Pollen	ND	Trace	Trace	1–2 %
Fungal, mold, biofilm	70–80 %	60–80 %	Trace	Trace
Soil minerals	15–25 %	10–20 %	90–95 %	90–95 %
Soot (including char)	Trace	ND	Trace	Trace
Flyash	ND	Trace	Trace	Trace
Glass fibers	ND	ND	Trace	ND
Plant fragments	ND	Trace	1–5 %	Trace
Paint	5–10 %	5–10 %	Trace	ND
Construction debris	ND	ND	ND	ND
Insect parts	ND	ND	ND	Trace
Starch	ND	Trace	ND	ND
Rust/metal flakes	ND	Trace	Trace	Trace
Rubber	ND	ND	Trace	Trace
Coal/coke	ND	ND	ND	ND
Catalyst	Trace	Trace	Trace–1 %	Trace

TABLE 3—*Average surface particulate composition by year.*

Particulate components	1998	1999	2000	2001	2002	2003	2004	
Pollen	0 %	0 %	3 %	1 %	NA	1 %	1 %	9 %
Fungal, mold, biofilm	2 %	5 %	3 %	8 %	NA	7 %	3 %	13 %
Soil minerals	23 %	18 %	40 %	33 %	NA	35 %	46 %	28 %
Soot (including char)	58 %	50 %	36 %	37 %	NA	32 %	24 %	7 %
Flyash	0 %	0 %	0 %	0 %	NA	0 %	0 %	0 %
Glass fibers	0 %	0 %	0 %	0 %	NA	0 %	0 %	2 %
Plant fragments	3 %	4 %	6 %	4 %	NA	10 %	8 %	7 %
Paint	0 %	11 %	1 %	3 %	NA	2 %	2 %	6 %
Construction debris	15 %	7 %	11 %	13 %	NA	9 %	0 %	1 %
Insect parts	0 %	0 %	0 %	1 %	NA	0 %	0 %	0 %
Starch	1 %	0 %	0 %	1 %	NA	0 %	2 %	0 %
Rust/metal flakes	1 %	0 %	0 %	0 %	NA	1 %	1 %	4 %
Rubber	0 %	0 %	1 %	1 %	NA	0 %	2 %	5 %
Coal/coke	0 %	0 %	0 %	0 %	NA	1 %	2 %	5 %
Catalyst	1 %	1 %	0 %	0 %	NA	1 %	1 %	1 %
Carbon black	14 %	8 %	8 %	2 %	NA	8 %	2 %	1 %
Total No. samples	2	13	17	6	0	11	73	39
Particulate components	2006	2007	2008	2009	2010			
Pollen	2 %	2 %	2 %	0 %	8 %			
Fungal, mold, biofilm	14 %	12 %	12 %	74 %	23 %			
Soil minerals	31 %	33 %	43 %	22 %	44 %			
Soot (including char)	3 %	3 %	2 %	0 %	2 %			
Flyash	0 %	0 %	0 %	0 %	0 %			
Glass fibers	2 %	1 %	0 %	0 %	0 %			
Plant fragments	16 %	14 %	11 %	2 %	11 %			
Paint	8 %	8 %	13 %	1 %	3 %			
Construction debris	14 %	9 %	2 %	0 %	1 %			
Insect parts	1 %	0 %	1 %	0 %	0 %			
Starch	1 %	2 %	1 %	0 %	1 %			
Rust/metal flakes	2 %	6 %	3 %	0 %	1 %			
Rubber	1 %	2 %	2 %	1 %	1 %			
Coal/coke	0 %	0 %	0 %	1 %	1 %			
Catalyst	0 %	1 %	1 %	0 %	1 %			
Carbon black	1 %	1 %	0 %	0 %	0 %			
Total No. samples	34	28	165	5	204			

minerals, soot, fungal (biofilms), and plant fragments were found to be significant contributors to the surface particulate. There was considerable variation from location to location for the particle types. Table 4 shows mean, standard deviation, minimum, maximum, and median values for the components of fungal material, soil minerals, soot particulate, and plant fragments. There is a distinct trend in the soot data where earlier years show higher average amount of

TABLE 4—*Study properties: Surface particulate average statistics (statistics versus year).*

		Fungal material							Soil minerals				
No. of samples	Year	Mean	STD	Min.	Max.	Median	No. of samples	Year	Mean	STD	Min.	Max.	Median
2	1998	2	1	1	2	2	2	1998	23	11	15	30	23
13	1999	5	7	0	25	3	13	1999	18	18	0	50	15
17	2000	3	7	0	30	1	17	2000	40	22	3	85	30
6	2001	8	13	1	35	3	6	2001	33	14	15	50	30
0	2002	0	0	0	0	0	0	2002	0	0	0	0	0
11	2003	7	13	0	40	1	11	2003	35	36	0	80	30
73	2004	3	10	0	80	1	73	2004	46	25	0	95	50
39	2005	13	21	0	100	4	39	2005	28	23	0	70	25
34	2006	14	17	0	50	5	34	2006	31	23	1	85	30
28	2007	12	21	1	85	3	28	2007	33	28	0	90	25
165	2008	12	19	0	97	3	165	2008	43	26	1	93	50
5	2009	74	41	1	93	93	5	2009	22	38	1	90	7
204	2010	23	30	0	100	3	204	2010	44	27	0	95	50

		Soot particulate							Plant fragments				
No. of samples	Year	Mean	STD	Min.	Max.	Median	No. of samples	Year	Mean	STD	Min.	Max.	Median
2	1998	58	39	30	85	58	2	1998	3	0	3	3	3
13	1999	50	37	0	95	50	13	1999	4	6	0	15	1
17	2000	36	29	3	87	30	17	2000	6	5	0	15	3
6	2001	37	29	1	70	50	6	2001	4	5	0	15	3
0	2002	0	0	0	0	0	0	2002	0	0	0	0	0
11	2003	32	39	0	90	4	11	2003	10	10	0	30	8

TABLE 4—*Continued*

Soot particulate							Plant fragments						
No. of samples	Year	Mean	STD	Min.	Max.	Median	No. of samples	Year	Mean	STD	Min.	Max.	Median
2	1998	58	39	30	85	58	2	1998	3	0	3	3	3
73	2004	24	21	0	97	18	73	2004	8	9	0	32	3
39	2005	7	12	0	52	2	39	2005	7	10	0	50	4
34	2006	3	6	0	25	1	34	2006	16	11	0	45	15
28	2007	3	3	0	13	3	28	2007	14	14	1	50	7
165	2008	2	4	0	50	1	165	2008	11	14	0	90	7
5	2009	0	0	0	1	0	5	2009	2	2	1	5	1
204	2010	2	7	0	98	1	204	2010	11	14	0	99	7

carbon soot in the surface samples. This is also true for the particles consistent with carbon black. There were individual samples in the early years that showed significant amounts of carbon black at some properties near the carbon black plant (within 1/2 mile). However, as shown in Fig. 17, there was a significant decrease in the level of surface soot that occurred in about 2005.

The usefulness of applying the combination of the mandatory TEM section and the non-mandatory PLM section of D6602 was illustrated by the analysis of a set of 48 samples collected over a two day period in March of 2005 by one sampling company. Fifteen of the samples were collected at sites several miles from the carbon black plant. The samples were originally sent to a laboratory to be analyzed only by the mandatory TEM section of D6602. The laboratory reported that all the samples were positive for carbon black. MVA Scientific Consultants performed a more complete analysis of splits of the 48 samples using both the TEM and PLM procedures in ASTM D6602 and found that the samples contained extremely high levels of carbon black material (in some cases 100 % of the particulate was carbon black). These high levels are very unusual for environmental samples away from a carbon black plant where other normal environmental types of particles are usually mixed in. The high levels were not replicated in additional sample collections done in 2006, 2007, and 2008 at the same locations by different collectors. The samples collected in 2006, 2007, and 2008 had carbon black levels that ranged from below detection to 8 %. Since no other multiple sample set collected from 1998–2010 showed such consistently high levels of carbon black, it is most likely that the sample set was either contaminated or intentionally adulterated.

Two individual samples of the 2005 set of 48 illustrate the unusual nature of the sample set. One sample collected on 03/04/2005 at a site approximately 2 and 1/4 miles south of the carbon black plant plant contained 99 % carbon black. Another sample collected on the same day (03/04/2005) at a site approximately 2 and 1/4 miles north of the carbon black plant contained 80–90 %

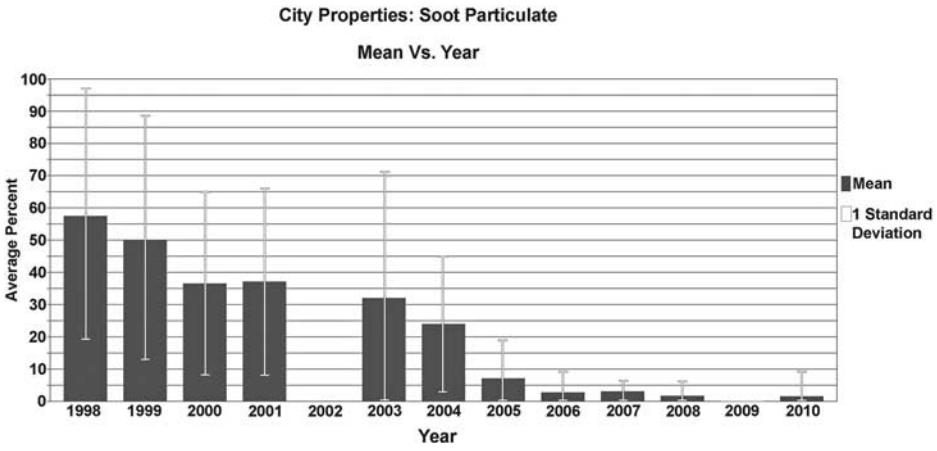

FIG. 17—*Histogram showing the average percentage of soot over the years in surface samples. Uncertainty bars are shown at one standard deviation.*

carbon black. It is totally inconsistent with the weather patterns of the day and the physics of dispersion of particulate that both of these samples nearly 5 miles apart in opposite directions could be composed almost entirely of carbon black from the carbon black plant facility.

Further inconsistencies were found in additional samples taken by a second sampling company, on the same day as the 2005 set of 48, but closer to the carbon black facility. Of the 4 samples collected by the second collector only one contained a trace amount of aciniform soot consistent with carbon black. It is not reasonable that a site close to the carbon black plant would have only a trace amount of carbon black while sites further away in the same direction on the same day would have nearly 100 % carbon black.

In addition to the surface particle components listed in Table 3, some dark particles were found that would not normally be present in environmental surface samples from other cities. Spent fluid catalytic cracking catalyst particles that could only have come from a petroleum refinery were found in 51 % of the samples. Depending upon when in the process the catalyst particles are emitted, they may be whitish, gray, or black. These particular particles were uniquely source identified by their appearance in PLM and their lanthanum content as determined by SEM x-ray analysis. Lanthanum is used to stabilize the zeolite cage structure for petroleum catalytic cracking. Black petroleum coke particles were found in 44 % of the samples. Petroleum coke can be differentiated from coal particles that might be released from passing trains by SEM x-ray analysis [39]. Petroleum coke contains only sulfur in addition to carbon and oxygen while bituminous coal particles contain a variety of elements calcium, silicon, and magnesium. Petroleum coke is a product of the refinery and stored on the refinery property in large piles until it is loaded with open shovel vehicles into trucks and train cars.

Discussion

There were a number of instances where the darkening on a surface was caused by fungal growth or obvious biofilms. In some of these cases, a trace amount of particulate that was consistent with aciniform soot or carbon black was also found (by TEM). It was clear that although the sample was positive for aciniform soot or carbon black, the real cause of the darkening was the fungal growth. It is concluded from a review of all the data that most current surface darkening situations are caused by non-industrial sources.

It is important to consider the semi-quantitative nature of the D6602 analysis. The particulate that make up a surface sample range from living biological organisms to inorganic matter in sizes that range from millimeters (some plant fragments) to nanometers (some soot particles). Because of the very diverse nature of the particulate that must be taken into consideration when evaluating a surface-darkening event, truly quantitative analysis is not possible at this time. While there were some situations where over 90 % of the particles in a sample were clearly of one classification and therefore the obvious darkening agent, it is in consideration of the multiple analysis results that information about the overall surface particulate situation is attained.

The USEPA uses a variety of methods to estimate the primary particulate emissions data for the traditionally inventoried anthropogenic source categories: (1) "Fuel combustion," which includes emissions from coal-, gas-, and oil-fired power plants and industrial, commercial, and institutional sources, as well as residential heaters and boilers; (2) "Other industrial processes," which includes chemical production, petroleum refining, metals production, and processes other than fuel combustion; (3) "On-road vehicles," which includes cars, trucks, buses, and motorcycles; and (4) "Non-road vehicles and engines," such as farm and construction equipment, lawnmowers, chainsaws, boats, ships, snowmobiles, aircraft, and others. For some years the USEPA also estimates the effects of natural sources, such as agriculture and forestry, wildfires and managed burning, and fugitive dust from paved and unpaved roads. Biogenic emissions and emissions from natural sources, such as plants and trees, are estimated using the Biogenic Emissions Inventory System Model, Version 3.12, with data from the Biogenic Emissions Landcover Database and annual meteorological data [40]. The data presented here for the microscopic analysis of surface particulate provide some direct information about the components of the surface material. The semi-quantitative data determined using the ASTM D6602 Standard is useful in understanding the overall picture of particulate deposition and biofilm growth that results in the accumulation of particles on surfaces.

Conclusions

The results of a multiyear study of atmospheric particulate material deposited on residential homes, city buildings and monuments indicates the value of the ASTM standard D 6602, "Standard Practice for Sampling and Testing of Possible Carbon Black Fugitive Emissions or Other Environmental Particulate, or Both." The use of the mandatory TEM section was able to identify the manufactured carbon black particulate and to distinguish it from other aciniform and non-aciniform carbon particles. The use of the non-mandatory sections involving light microscopy and SEM x-ray analysis yielded a very good semi-quantitative understanding of the composition and relative amounts of various other types of atmospheric particulate material. The use of the non-mandatory PLM and SEM analysis also allowed the identification of tracer particles that are unique markers for a particular source. In this case, the catalyst particles found in the environmental samples were a tracer for emissions from the refinery.

Long term decreases in the quantity of aciniform and non-aciniform soot were found in the samples collected from 1998 through 2010. A particularly dramatic decrease in all types of soot particulate was noted in mid 2005 and continued thereafter. This decrease in soot coincided with upgrades in particulate control technologies at the two major industrial sites in the city, a refinery, and a carbon black plant. Other surface agents such as fungal growth were identified and found to be a significant cause of surface darkening.

The use of a well studied and documented ASTM Standard Practice allowed multiple analytical laboratories to obtain similar results on split samples. Semi-quantitative analyses had a repeatability of approximately 15 % and reproducibility among different analysts of approximately 30 %.

Disclosure

During the analysis of the outdoor surface samples collected in a city in Oklahoma and surrounding areas, the authors were under contract to the carbon black plant to provide an independent, scientific assessment of the darkening on properties in the surrounding area. They had the right to publish the data regardless of how the results came out. One of the authors, J.R.M., has worked for both plaintiffs and defendants in lawsuits involving possible carbon black property contamination.

References

[1] U.S.C. 7401-7626, 1990, *National Ambient Air Quality Standards, Clean Air Act*, U.S. Environmental Protection Agency.

[2] EPA 452/P-10-003, 2010, *Policy Assessment for the Review of the Particulate Matter National Ambient Air Quality Standards, First External Review Draft*, U.S. Environmental Protection Agency, Office of Air Quality Planning and Standards, Research Triangle Park, NC.

[3] ASTM D3274-09e1, 2009, "Evaluating Degree of Surface Disfigurement of Paint Films by Microbial (Fungal or Algal) Growth or Soil and Dirt Accumulation," *Annual Book of ASTM Standards*, Vol. 06.01, ASTM International, West Conshohocken, PA.

[4] ASTM STP D4610, 2004, "Determining the Presence of and Removing Microbial (Fungal or Algal) Growth on Paint and Related Coatings," *Annual Book of ASTM Standards*, Vol. 06.01, ASTM International, West Conshohocken, PA.

[5] ASTM STP D6602-03b(2010)e1, 2010, "Sampling and Testing of Possible Carbon Black Fugitive Emissions or Other Environmental Particulate, or Both," *Annual Book of ASTM Standards*, Vol. 09.01, ASTM International, West Conshohocken, PA.

[6] Millette, J. R., Turner, W., Hill, W. B., Few, P., and Kyle, J. P., "Microscopic Investigation of Outdoor 'Sooty' Surface Problems," *Environ. Forensics*, Vol. 8, 2007, pp. 37–51.

[7] Turner, W. L., Millette, J. R., Boltin, W. R., and Hopen, T. J., "A Standard Approach to the Characterization of Common Indoor Dust Constituents," *Microscope*, Vol. 53(4), 2005, pp. 169–177.

[8] Millette, J. R., Brown, R. S., and Hill, W. B., "Using Environmental Forensic Microscopy in Exposure Science," *J. Exp. Sci. Environ. Epidemiology*, Vol. 18, 2008, pp. 20–30.

[9] McCrone, W. C., Delly, J. G., and Palenick, S. J., *The Particle Atlas*, 2nd ed., Vol. 2, Arbor Science, Ann Arbor, 1979.

[10] Palenik, S. J., "The Determination of Geographical Origin of Dust Samples," *The Particle Atlas*, 2nd ed., Vol. 5, W. C. McCrone, J.G. Delly, S. J. Palenick, Eds. Ann Arbor Science, Ann Arbor, 1979, pp. 1347–1361.

[11] Cassens, D. L. and Feist, W. C., "Discoloration of House Paint—Causes and Cures," U.S. Department of Agriculture and Cooperative Extension Services of the North Central Region, NCR Extension Publication, Vol. 134, 1988.

[12] Gorbushina, A. A., Krumbein, W. E., Hamman, C. H., Panina, L., Soukharjevski, S., and Wollenzien, U., "Role of Black Fungi in Color Change and Biodeterioration of Antique Marbles," *Geomicrobiol. J.*, Vol. 11, 1993, pp. 205–220.

[13] Graves, W. J., "A Mineralogical Soil Classification Technique for the Forensic Scientist," *JFSCA*, Vol. 24, 1979, pp. 323–338.

[14] Hawksworth, D. L., Kirk, P. M., Sutton, B. C., and Pegler, D. N., *Ainsworth and Bisby's Dictionary of the Fungi*, CABI International, Oxon, United Kingdom, 1995, p. 616.
[15] Faegri, K., Iversen, J., and Waterbolk, H. T., *Textbook of Pollen Analysis*, Hafner Publishing Company, New York, 1964, p. 237.
[16] Farr, D., Bills, G., Chamuris, G., and Rossman A., *Fungi on Plants and Plant Products in the United States*, American Phytopathological Society Press, St. Paul, 1989, p. 1152.
[17] Hopen, T. J., "The Value of Soil Evidence," *Trace Evidence Analysis*, M. M. Houck, Ed., Elsevier Academic, Boston, 2004, pp. 105–122.
[18] Krumbein, W. C. and Pettijohn, F. J., *Manual of Sedimentary Petrography*, Appleton-Century-Crofts, New York, 1938, pp. 439–441.
[19] May, E., Lewis, F. J., Pereira, S., Taler, S., Seaward, M. R. D., and Allsopp, D., "Microbial Deterioration of Building Stone—A Review," *Biodeterioration Abstr.*, Vol. 7, 1993, pp. 109–123.
[20] Folk, R. L., "A Comparison Chart for Visual Percentage Estimation," *J. Sediment. Petrol.*, Vol. 21(1), 1951, pp. 32–33.
[21] Travis, R. B., "Classification of Rocks," *Q. Colorado School Mines*, Vol. 50(1), 1955, p. 8.
[22] Terry, R. D. and G. V. Chilingar, "American Geological Institute Data Sheet 6," *J. Sediment. Petrol.*, Vol. 25, 1955, pp. 229–234.
[23] Reid, J. C., "Comparison Chart for Estimating Volume Percentages of Constituents in Rocks and Concentrates in the Range of 1.0 to 0.1 Volume Percent," *Am. Miner.*, Vol. 70, 1985, pp. 1318–1319.
[24] Baccelle, L. and Bosellini, A., "Diagrammi per la stima visiva della composizione percentuale nelle rocce sedimentary," *Sci. Geol. Paleontol., Ann. Univ. Ferrara, N. Ser.*, Vol. IX:1, 1965, pp. 59–62.
[25] EPA-600/M4-82-020, *Test Method: Interim Method for the Determination of Asbestos in Bulk Insulation Samples*, U.S. Environmental Protection Agency, 1982.
[26] NIOSH 9002, "Asbestos (bulk) by Polarized Light Microscopy (PLM) — Method 9002," Issue 2, *NIOSH Manual of Analytical Methods*, 4th ed., National Institute of Occupational Safety and Health, U.S. Department of Health and Human Services, 1994, p. 94.
[27] Millette, J. R. and Turner, W. L., Jr., "Visual Estimation in the Analysis of Surface Particulate by Microscopy," *Microscope*, Vol. 58(2), 2010, pp. 65–68.
[28] Harvey, B., Doorn, S. S., Ennis, J. T., Greene, L. C., Winstead, W. G., Gerald, M., and Robbins, E.E., "American Industrial Hygiene Association Bulk Asbestos Proficiency Analytical Testing Program. Final Report to Laboratories—Test round A73-407," Report No. RTI/04562/73L-F, 2008.
[29] Clague, A. D. H., Donnet, J. B., Wang, T. K., and Peng, J. C. M., "A Comparison of Diesel Engine Soot with Carbon Black," *Carbon*, Vol. 37, 1999, pp. 1553–1565.
[30] Goldberg, E. D., *Black Carbon in the Environment: Properties and Distribution*, Wiley, New York, 1985, p. 187.
[31] Hess, W. M. and Herd, C. R., "Microstructure, Morphology and General Physical Properties," *Carbon Black: Science and Technology*, 2nd ed., J. Donnet, R. C. Bansal, and M. Wang, Eds., Marcel Dekker, New York, 1993, pp. 89–173.
[32] Huffman, G. P., Huggins, F. N., Shah, N., Huggins, R., Linak, W. P., Miller, C., Pugmire, R. J., Meuzelaar, H. L. C., Seehra, M. S., and Manivannan, A., "Characterization of Fine Particulate Matter Produced by Combustion of Residual Fuel Oil," *J. Air Waste Manage. Assoc.*, Vol. 50(7), 2000, pp. 1106–1114.
[33] Kostiuk, L. W., Johnson, M. R., and Thomas, G. P., "University of Alberta Flare Research Project Final Report Nov. 1996 – Sept. 2004," University of Alberta, Department of Mechanical Engineering, 2004, pp. 162–180.

[34] Medalia, A. I. and Rivin, D., "Particulate Carbon and Other Components of Soot and Carbon Black," *Carbon*, Vol. 20, 1982, pp. 481–492.
[35] Millette, J. R. and Few, P., "Indoor Carbon Soot Particles," *Microscope*, Vol. 46(4), 1998, pp. 201–206.
[36] Stevenson, R., "The Morphology and Crystallography of Diesel Particulate Emissions," *Carbon*, Vol. 20(5), 1982, pp. 359–365.
[37] Laboratory Standards and Technologies, 2010, www.carbonstandard.com/ (currently seven grades of carbon blacks that have been approved by ASTM Committee D24 are available) (Last accessed 1 August 2011).
[38] NIST SMR 2975, "Diesel Particulate Matter," *U. S. National Institute of Standards and Technology*.
[39] Millette, J. R., Brown, R. S., Kyle, J. P., Turner, W. L., Jr., Hill, W. B., and Boltin, W. R., "Distinguishing Coal, Coke and Other Black Particles," *Microscope*, Vol. 57(2), 2009, pp. 51–57.
[40] U.S. Environmental Protection Agency, NEI (National Emission Inventory) http://cfpub.epa.gov/eroe/index.cfm (Last accessed 1 August 2011).

Reprinted from JAI, Vol. 8, No. 10
doi:10.1520/JAI103656
Available online at www.astm.org/JAI

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Surface Characterization of Replicate Wood Surfaces for Cleaning Studies

ABSTRACT: A need exists to develop realistic surfaces, with differing degrees of surface roughness, to standardize methods for sampling and cleaning of environmental contaminants. This study reports on development and characterization of a novel means to create reproducible wood surfaces that are approximately 7×10 cm^2 and have differing degree of surface roughness. Three unique specimens were prepared by creating 3-dimensional digital-scans of actual wood surfaces found in three St. Louis homes. These surfaces had distinct depressions that likely arose from tool marks. A reverse image die of each specimen was made from high density plastic. Replicates of the three specimens were created by placing a die and wood veneer section into a rolling-press leaving the original topographies gathered from the homes. These specimens were used in a separate study to evaluate cleaning leaded dust using wipe or vacuuming treatments. The separate cleaning studies found no difference in cleaning due to the effects of the different surfaces. An in-depth examination of the three specimens was held to determine if differences in surface roughness of the specimens

Manuscript received November 30, 2010; accepted for publication July 20, 2011; published online August 2011.

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Cite as: Lewis, R. D., Kennedy, J., Andruskiewicz, M., Brown, C. A. and Condoor, S., "Surface Characterization of Replicate Wood Surfaces for Cleaning Studies," *J. ASTM Intl.*, Vol. 8, No. 10. doi:10.1520/JAI103656.

explained removal of lead from its surface. A method using optical, 3-D laser profilometry was used to determine if there were statistical differences between the surface roughness of the three specimens and within the specimens themselves. Area-scale analyses were made to calculate relative areas as a function of scale as a way of characterizing the roughness. Statistical differences were found for surface roughness between each of three surface specimens at a scale where small particles, deposited in the cleaning studies may adhere to the wood. However, within the specimens, surface roughness, at the scale where fine particles may lodge, were not consistent inside or outside tool marks. Therefore, differences in surface roughness were found within and between surfaces but they may not be large enough or different enough for detection using simple cleaning methods. Specimens with larger differences in surface roughness may be needed to simulate differences in cleaning effectiveness. This work should be continued to understand the scales for the different interactions of particle deposition, adhesion, and removal toward development of standard reference surfaces with well-characterized textures.

KEYWORDS: surface, roughness, sampling, cleaning, wood, profilometry, laser

Introduction

Surface sampling research and the development of standards which rely on methods to quantitify measures of inorganic, organic, and viable contaminants on surfaces, could benefit from a range of standard-complex surfaces for deposition of standard reference contaminants [1,2]. The ASTM D7659-10, *Standard Guides for Surface Sampling of Metals and Metalloids for Worker Protection*, states that "consideration should be given to the expected means by which the material being sampled was deposited on the surface or *surface being sampled* [3]. The point is that both deposition mechanisms and *surface roughness* will affect the adhesion and removal of environmental contaminants during sampling and by inference, cleaning, since good cleaning must overcome the same forces of particle adhesion as sampling to get acceptable removal of contaminates. The ASTM 7144-05a, *Standard Practice for Collection of Surface Dust by Micro-vacuum, Sampling for Subsequent Metal Determination*, is intended for use on rough, textured surfaces [4]. This method, unlike wipe sampling methods, can remove particles from crevices of rough materials because it uses an air sampling pump and appropriate sampling nozzle for metal dust removal. The effectiveness of this method is good but the method was not evaluated on *standardized*, rough surfaces [5]. As new surface sampling methods are developed or existing surface sampling or cleaning methods are revised, a need exists to develop and characterize replicate surface-materials that are textured and represent real materials, such as wood flooring, for evaluating the effectiveness of a sampling or cleaning device.

Although custom surface materials have previously been produced in replicate for studying allergen retention on carpets, such surfaces have rarely been made and are not commonly available for study of particle deposition, sampling, and removal from real surfaces [6]. Nine separate ASTM standards on lead or metal sampling plus the United States Department of Housing and Urban Development (HUD) guidelines for removal of lead from residential settings were reviewed for this paper and none of these use replicate-surfaces to simulate how roughness affects deposition and removal efficiency of particles in the field [3,4,7–14].

A need arose to develop replicate wood specimens with differing degrees of surface roughness in order to simulate cleaning of lead dust from wood with different cleaning techniques, including wet wipes and electrostatic cloths [15,16]. Because window sills, window troughs, and floors are locations in a home that need to be cleaned and sampled prior to clearance during lead abatement [14] we looked for different yet highly textured, in situ specimens, in St Louis homes undergoing lead abatement, that could be duplicated and reproduced for a laboratory study of cleaning. The surfaces we found were from three different homes that had distinct impressions or depressions in them which likely arose from tool marks like hammers, nails, and grooves from saw cuts or other tools. The gross surface textures were thought to be difficult to wipe or clean and the three specimens seemed different enough from each other to warrant their use. However, no significant differences were found among the replicate-wood surfaces for cleaning of lead [15,16]. The finding of a lack of a significant main effect of surfaces to affect cleaning went contrary to previous work where wallpaper textures were found to be significantly different in their ability to hold lead dust than wood or linoleum surfaces [17]. We decided to investigate the surface roughness of the wood specimens to determine if in fact there were major differences in spite of the inability of these surfaces to show much difference in lead dust-cleaning experiments. One hypothesis we tested is that the fine scale topography of the wood inside depressions left by tools is different from the topography outside.

Some wipe sampling or cleaning methods may be adequate to remove fine particulate; for example, particles less than 75 μm, from holes or depressions in wood surfaces. Do the holes and depressions sometimes found in wood surfaces in homes lead to higher surface roughness that retains the fine dust or is the fine dust retained just as well outside the holes and depressions as within them? Conventional contact profilometry can establish the 2-dimensional contours of a surface, and can tell us if surfaces differ in the peaks and valleys of surfaces that we can often see. The laser profilometer can tell us whether differences exist in surface roughness at the size of the particles that may be interacting with the surface, at the micron level. By using optical methods, such as the laser profilometer, we can establish not only if the surfaces are indeed different from each other in their surface roughness but also if this difference is at the scale where particle adhesion may take place. To develop replicate-specimens for conducting wipe or cleaning studies or developing standards it is important to know what could be responsible for retaining the particles; such as differences in surface roughness across specimens or differences in surface roughness

within specimens due do the many ruts, holes, and grooves that can emerge on aged wood that has experienced tool work. For developing standards on cleaning or sampling surfaces where lead abatement is conducted, surfaces that might simulate the challenges of cleaning and wiping due to their apparent rough topography are needed.

The specific objectives of this paper are to report on the manufacture of the wood specimens and their surface roughness. It is thought that characterization of the surface roughness of coupon size materials such as the wood specimens in this study is an important first step for establishing standard reference materials for cleaning and sampling studies. The scope of the surface roughness characterization was limited to using area-scale analysis, a kind of scale-sensitive fractal analysis, to analyze measured topographies. Area-scale analysis shows how the area of a surface varies with the scale of observation and provides a characterization of the surface topography over the range of scales available in the measurement. Scale-sensitive fractal analysis [18] has been found useful for discriminating surfaces where other methods have failed [19]. In addition it has been found to correlate well with deposition of particles on carpets [20].

Methods

Wood Specimen Production

The materials used are listed below:

- 3D Rapid Prototyping Machine.
- Technovate Roller Mill.
- Ultra HighMolecular Weight Polyethylene Plastic—2.54 cm thick, cut to 7.6 x 10.2 cm molds (3).
- White Pine—3.2mm thick, cut to 7.6 × 10.2 cm pieces (300).
- Molding Clay.
- Plaster.
- Polyurethane Minwax® Superfast-Finish Compound.

Sections of flooring or window sills were collected. Clay (Sargent Art 22-4084, Hazelton, PA) impressions were collected from three wood locations in homes undergoing lead abatement: a nail-indented window sill, a heavily grooved hardwood floor, and a contoured window sill. Figure 1 illustrates a final, painted reproduction of the nail-indented windowsill as it sits on top of the original surface. Clay was pressed into each of these surfaces, filling a 7.6×10.2 cm^2 area.

Next, plaster negatives, approximately 7.6×10.2 cm^2 area, (Plaster of Paris, 53005, DAP, Baltimore, MD) were created from the clay impressions, and these negatives mimicked positive images of the original surface (Fig. 2). The plaster was scanned digitally using a 3D Scanner and Rapid Prototyping machine (LCI Signs and Ironworks, Pine, CO) and images were created of the original surfaces. The images were inverted, and the inverted images were cut from 2.54 cm thick, high density plastic to make dies, using the Rapid Prototyping Machine (Fig. 3).

FIG. 1—*Original nail-indented surface (dark wood) with its final painted (white) specimen, Number 1, on top.*

White pine was cut into 7.6 × 10.2 cm pieces, 3.2 mm thick. The pieces were aligned with high-density dies and run through a Technovate Roller Mill (T9013, Amatrol, Inc., Jeffersonville, IN), to create an accurate duplicate of the original hard surface (Fig. 4). White pine has a hardness of 569.56 NM (440 lbf)with Janka hardness test, (ASTM D1037) which corresponds to 18.68 MPa (1.7×10^4 PSI), and we used approximately 6 times that, applying 117.21 MPa (17k psi) [21]. One-hundred replicates were created for each surface type, for a total of 300 specimens. The entire wood surface was compressed somewhat by the roller, with the tool marks being more severely compressed. Specimens that were cracked, split, or warped were destroyed. The wood specimens were coated with a single layer of polyurethane (Minwax Fast-drying polyurethane, Upper Saddle River, NJ) or with three layers of white-latex paint and allowed to dry completely. Once dry, the specimens were ready to be coated with dust and tested. The uncoated specimens are depicted in Fig. 5. Specimens are: (1) nail-indented window sill, (2) Deep-grooved floor, and (3) Slightly contoured window sill. A blue circle was added to the figure in order to illustrate the tool mark. The height measurements from the tool mark were separated from the rest of the measurement by a process called thresholding, which is described later in the text.

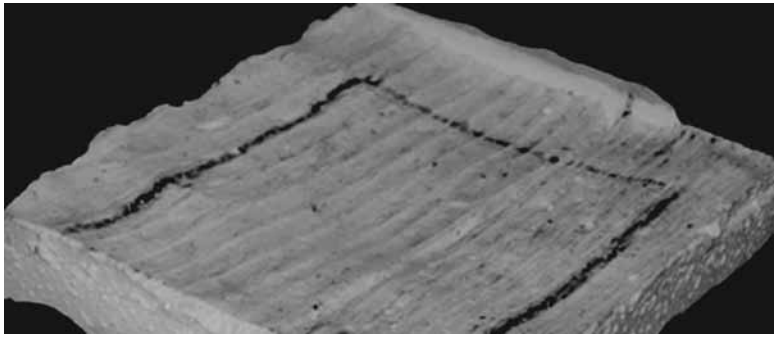

FIG. 2—*Plaster cast of heavily grooved hardwood floor (7.6 × 10.2 cm^2 area).*

FIG. 3—*High-density plastic dies of the three specimens (top to bottom: Spec 2, heavily grooved floor; Spec 1, nail-indented window sill, Spec 3, contoured window sill).*

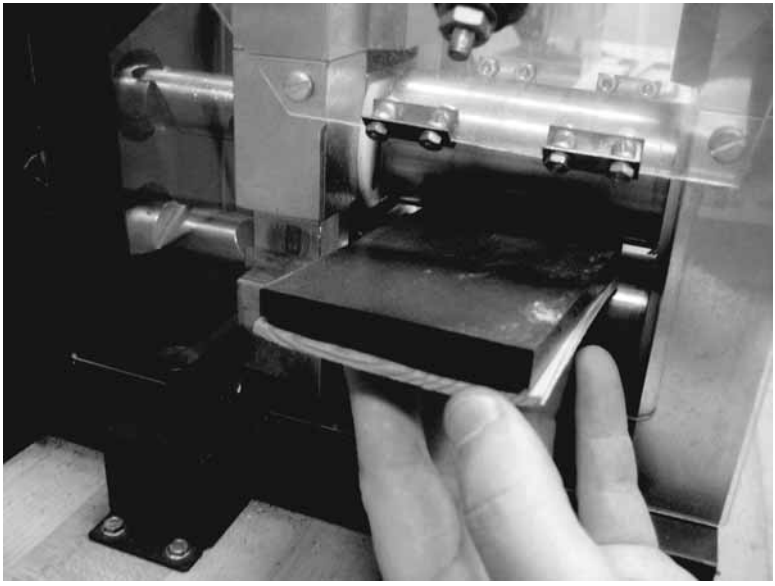

FIG. 4—*Duplicates of the wood specimens are made using a die in the Technovate Rolling Mill (photo shows specimen going into the press).*

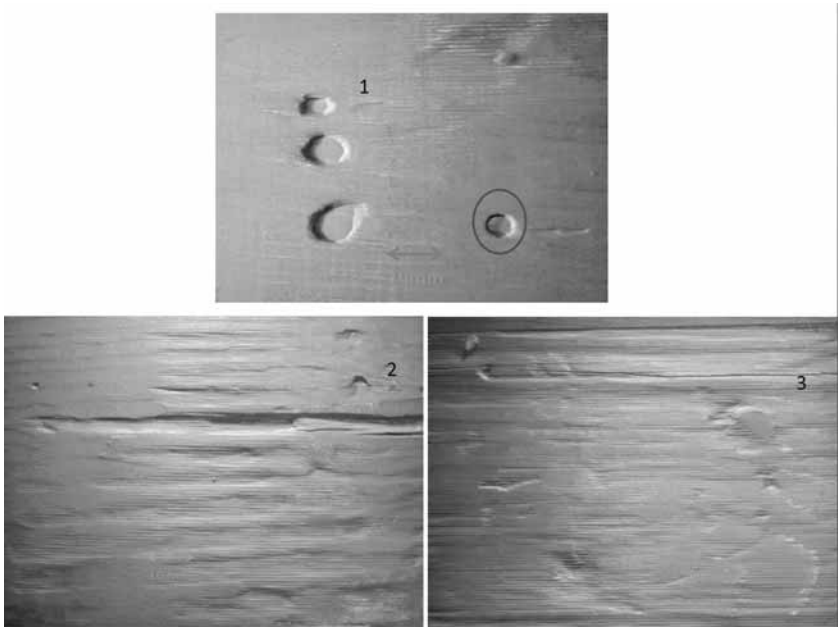

FIG. 5—*Test Specimens: (A) Specimen 1 – Nail-indentations from window sill, (B) Specimen 2- Deep grooves from hardwood floor, (C) Specimen 3 – Contoured Surface from window sill.*

Surface Topography Measurements

A UBM laser-profilometer (Ulrich Breitmeir Messtechnik, Ettlingen, Germany; distributed in the US by Solarius Development Co., Sunnyvale CA), and Keyence Laser Triangulation Sensor (Model LC2210, Keyence Corporation, Osaka, Japan), were used to measure the topographies of the surfaces of the wood specimens (Fig. 6). In this device the piece with the surface to be measured is secured to a stage that moves horizontally in x and y under a sensor that measures heights in z. The height sensor has a spot size of about 70 μm; however it is able to resolve height differences with spatial (x, y) dimensions less than 25 μm. Currently there is no US or ISO standard for determining the spatial height resolution of sensors of this kind of surface topographical measurement instrument. The sampling interval between measured heights was 10 μm in x and y and the measurement region was 10×10 mm. The measurements consist of 1001 height-profiles incremented in y, each profile containing 1001 heights ($z = z(x)$), forming a total measurement of over 1 000 000 heights on a regular horizontal grid in x and y (i.e., $z = z(x, y)$, meaning that the height z is a function of the position in x and y).

Three measurements were taken of each of the specimens away from the tool marks. This approach was used, separating the measurement inside and outside the tool mark, in order to see if the action of the tool has the same topography of the wood at a fine scale, below the relatively large scale of the

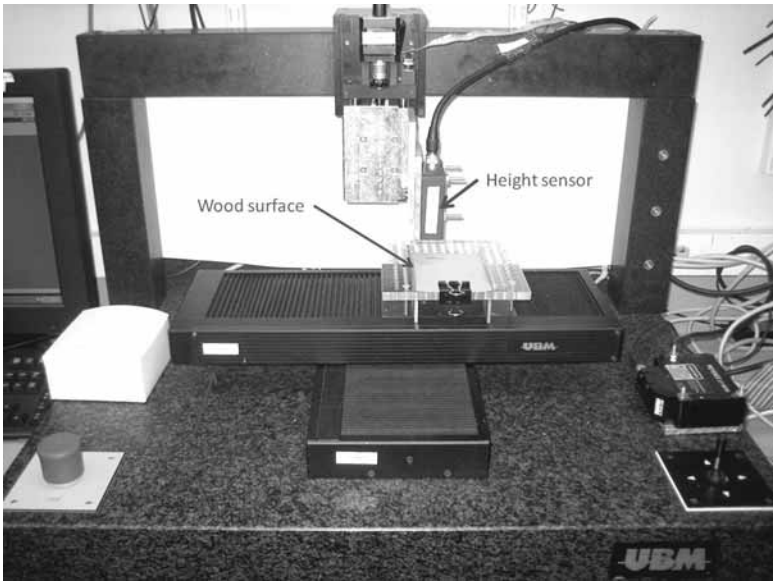

FIG. 6—*UBM scanning laser profiler consisting of a granite metrology frame with a Keyence Laser Triangulation Sensor (model LC2210) mounted on it to measure the heights; the surfaces to be measured are placed below the height sensor on stages that move in x and y to scan the surface under the height sensor.*

indentation caused by the tool. This facilitates testing the hypothesis that the tool has modified the roughness of the wood at a fine scale. Three other measurements were made of regions including depressed areas that were defined by tool marks. Each measurement contains about 1×10^6 measured elevations. The measurements taken away from the tool marks were divided into four adjacent measurements each with about a quarter of a million measured elevations. This was done in order to have separate measurements for better statistical comparison testing between the three wood specimens. The regions inside the tool marks were separated from those outside by thresholding, a process that uses a height threshold to separate the lower altitude measurements in the tool mark from the higher ones on the wood surface. The thresholding process essentially cuts the measurement into two parts horizontally. This way the depressed area made by the tool mark is in one part and the rest of the measurement is in the other part (Fig. 7).

Topographic Characterization

The topographic measurements were leveled and area-scale analyses [19] were made using Sfrax (www. Surfract.com). Area-scale analysis is a kind of scale sensitive fractal analysis. It is based on the principle that the area of a rough surface depends on the scale of observation or calculation. The calculation of the area is made with a virtual tiling algorithm [22]. The algorithm uses

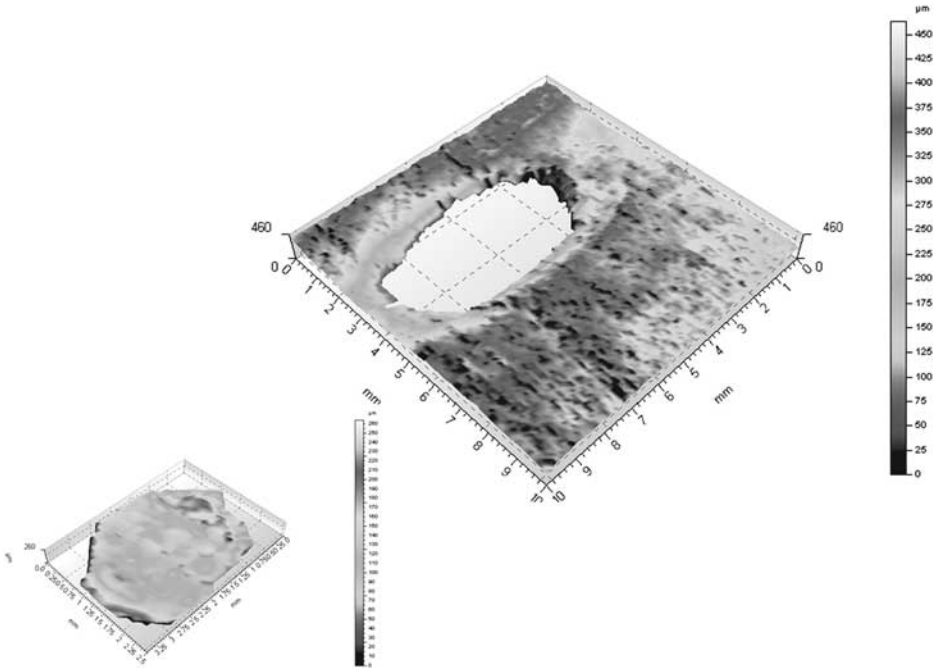

FIG. 7—*Surface measurement (specimen 1) with tool mark removed by thresholding, i.e., separating the lower elevations inside the depression caused by the tool mark through selecting a threshold in height.*

repeated tiling exercises (Fig. 8). The tiles have a certain, constant area in three dimensions, but with a shape that can vary within limits. The corners of the triangles are located on lines interpolated between the measured heights. The area of the triangle represents the scale of observation or calculation. The area of the triangle is varied systematically from exercise to exercise so that the range from the scale of the measured region to the sampling interval is covered.

At each areal scale (s) a relative area (RelA(s)) is calculated. The relative area at each scale is the ratio of the calculated area (CalA(s)) to the nominal area (NomA(s))

$$\mathrm{RelA(s)} = \mathrm{CalA(s)}/\mathrm{NomA(s)}$$

The calculated area at each scale is the number of triangles used in the tiling (N(s)) times their area, which is the scale (s)

$$\mathrm{CalA(s)} = \mathrm{N}^{*}\mathrm{s}$$

The nominal area is the large scale, x-y, or projected area, of the region included in the tiling exercise at that scale. The algorithm has been described in detail [18] and its use is found in the technical literature [19]. The result of the area-

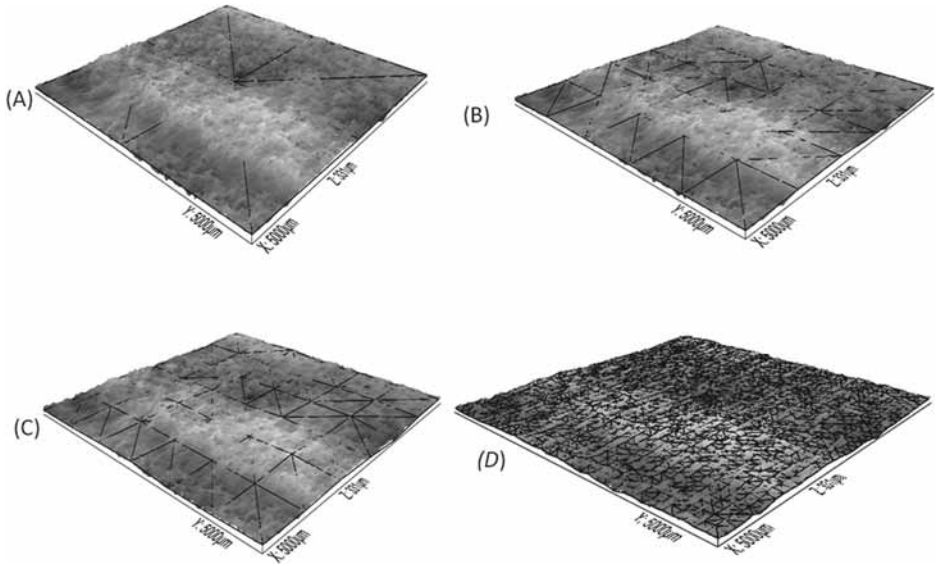

FIG. 8—*Virtual tiling exercise for determining relative areas as function of scale on sections 5000 × 5000 μm. The equations for calculating relative area are in the text. The max peak to valley distance for this measurement is 331μm. The number of triangular tiles, the scale, or area of the triangular tiles and the relative areas are: (A) 8 tiles, scale = 30|075|200 μm², RelA = 1.00016; (B) 50 tiles, scale = 490|050 μm², RelA = 1.00130; (C) 72 tiles, scale = 336|200 μm², RelA = 1.00384; (D) 2242 tiles, scale = 11|250 μm², RelA = 1.03470.*

scale analysis is an area-scale plot, which is a log-log plot of the relative area versus the scale.

Scale-based calculations of the area of rough surfaces are useful descriptions of the geometry of rough surfaces for correlating with behavior and with processing [23]. The area of a rough surface is not unique; rather it changes with the scale of observation. When a rough surface is examined more closely, topographic details become evident which were not obvious at larger scales. These topographic details tend to increase the apparent area of the surface. As the area increases the inclinations on the surface increase as do the opportunities to create bond with the surface. It is reasonable to propose a hypothesis that surfaces that have larger relative areas, at the scale where particles are retained, will have a greater capacity to retain these particles [20]. The scales of interaction with a rough surface, like retention, can be determined experimentally [23].

To test for differences in the surface measurements a modified F-test, including the mean and standard deviation, is used at each scale to compare the relative areas of the different wood at that scale [24]. The results of these analyses are mean square ratios, corresponding to the scales of the relative areas used in the test. The mean square ratios are plotted as a function of the

corresponding scales. The minimum, mean square ratio for a certain level of confidence in the ability to discriminate can be shown on the plot.

Results

The results of the surface characterization of the three wood specimens are described in several figures. Figure 9 shows a representation of a measurement of the wood surfaces on the scanning laser profiler, which would subsequently be divided into four regions for statistical analysis. Figure 7, as previously described, shows a representation of a measurement taken from Specimen 1. The nail-indented window sill included a tool mark (see Fig. 5, blue circle) that was removed by thresholding. The removal separates the topographies that may have been modified by the tool from those that were not influenced by the tool. This facilitates testing the hypothesis that the action of compressing the wood with the tool has modified the topography at a fine scale. Similar procedures were followed for tool marks on specimens 2 and 3.

Figure 10 shows area-scale plots of the 12 measurements from two of the wood surfaces (specimens 1 and 3). Notice in Fig. 10 that at the largest scales the relative areas are close to one, indicating that at those scales the surfaces

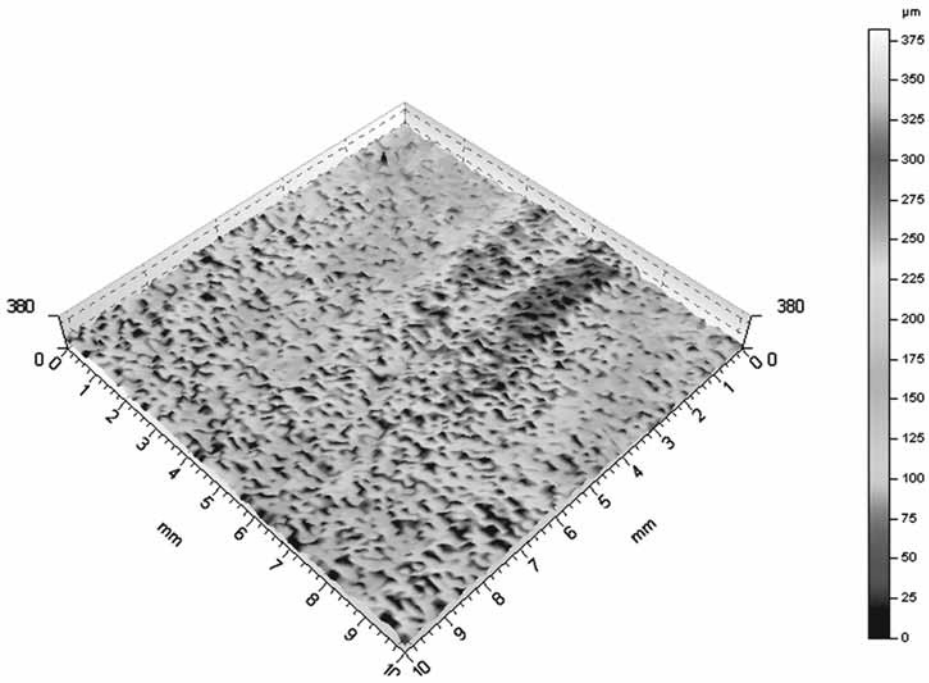

FIG. 9—*Elevation map of specimen 3, rendering a measurement made over a field of 10 × 10 mm with 1|002|001 measured elevations, or heights, on a regular 1001 × 1001 horizontal grid.*

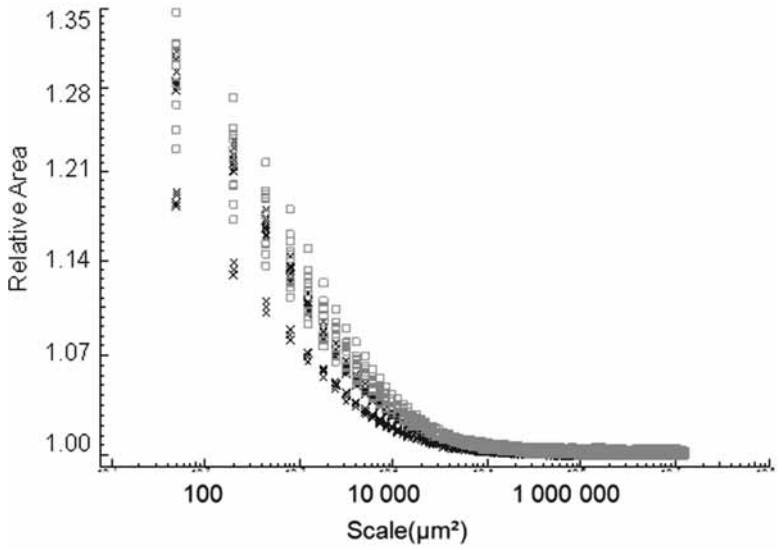

FIG. 10—*Area-scale plots of 12 measurements for comparing specimens 1 and 3 (specimen 1 is represented by x, specimen 3 is represented by squares).*

are essentially smooth. Below scales of about 10 000 μm^2, the relative areas have deviated clearly from one and at these finer scales the surfaces are essentially rough. This is the smooth-rough crossover scale, i.e., the scale above which the surface appears to be smooth and below which, i.e., at finer scales, it appears to be rough. At the finest scales the relative areas vary from about 1.18 to 1.35. It is not known what relative areas should be typical for wood. This may be the first analysis of wood topographies of this type. Milled metal surfaces can have relative areas between 1.005 and 1.02 [23]. In general specimen 3 (contoured surface) has larger relative areas than specimen 1 (nail-indented surface).

In the plots of relative area and of the mean square ratio (MSR) plots versus scale, the x-axis represents the areal scale (Figs. 10–16). This areal scale on the x-axis is the same areal scale as the area of the triangle used in the tiling exercises to determine the calculated area of the measured surface topography [18] [19]. In the algorithm for the tiling exercises the triangles are inclined to the nominal horizontal, or x-y, plane during the tiling exercises; therefore the areal scales, as used here, have components in horizontal (x, y) and vertical (z) directions [25].

Differences in surface roughness across specimens

The mean square ratio is a parameter that results from the modified F-test used to test for the discrimination confidence [21]. The confidence level is determined from the df [21] and is indicated on the plots. The mean square ratios from the F-test for testing for discrimination of specimens 1 and 2 and 1 and 3, by relative areas as a function of scale, are shown respectively as Figs. 11 and 12. A high

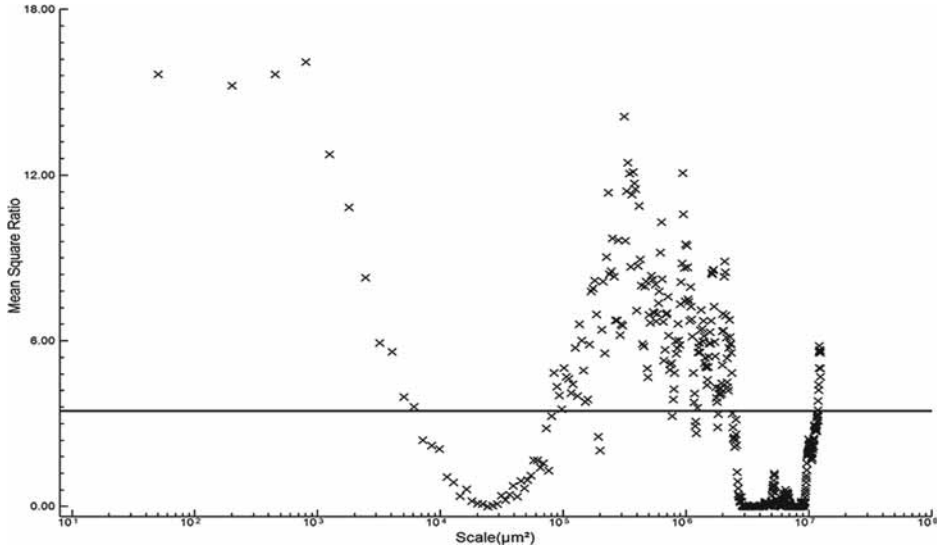

FIG. 11—*Mean square ratios (MSR, which is unitless) versus scale from an F-test of relative areas comparing specimens 1 versus 2; horizontal line is the minimum MSR for 95 % confidence that the surfaces are different over the represented scales.*

degree of ability to discriminate with 95 % confidence is indicated for surfaces 1 and 2 below 5000 μm^2 and between 100 000 to about 2 000 000 μm^2. This is indicated because over this scale range the MSR exceeds the threshold based on the df in the test [21]. For Fig. 12, discrimination of surfaces 1 and 3 are found from

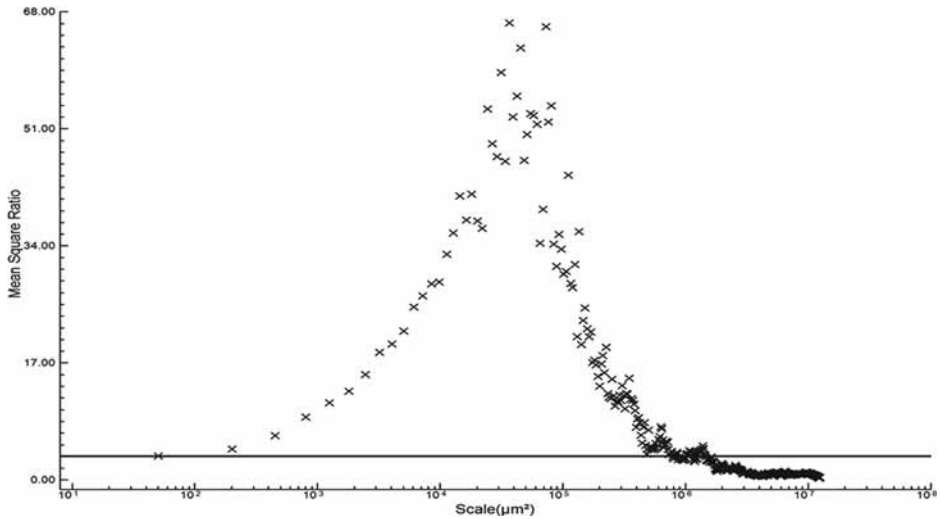

FIG. 12—*Mean square ratios (MSR) versus scale from an F-test of relative areas, comparing specimens 1 versus 3; horizontal line is the critical MSR for 95 % confidence.*

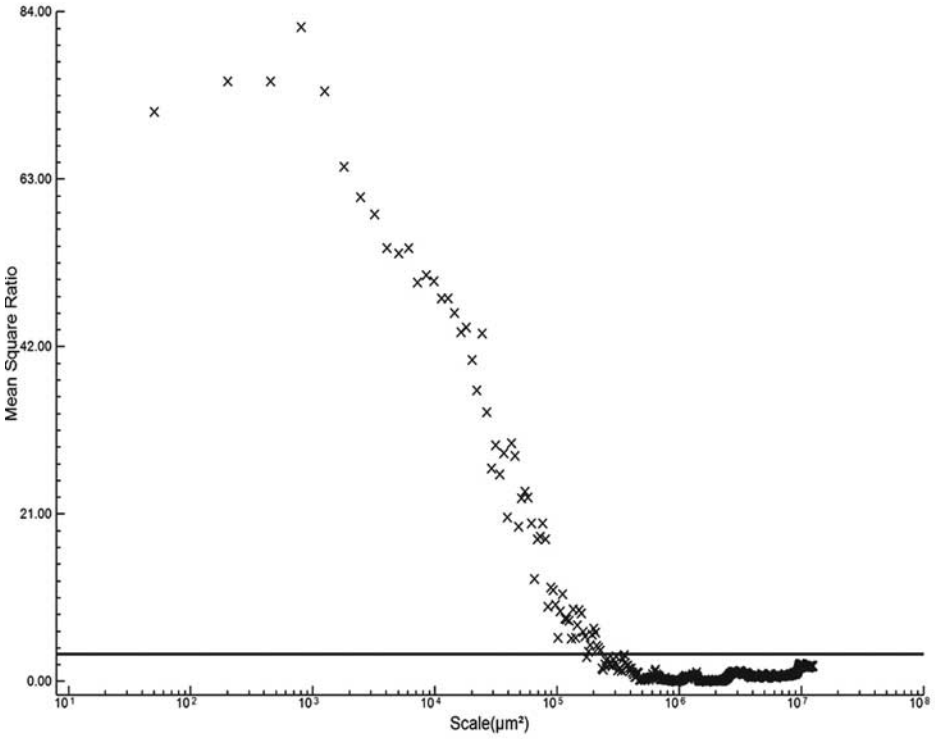

FIG. 13—*Mean square ratios (MSR, which is unitless) versus scale from an F-test of relative areas, comparing specimens 2 versus 3; horizontal line is the critical minimum MSR for 95 % confidence that the surfaces are different over the represented scales.*

about 200 μm^2 to about 400 000 μm^2. These scales correspond to those in the area-scale plot where the 12 measures of relative areas (Fig. 10) on each surface are well grouped.

The discrimination scales between surfaces 2 and 3 are shown in Fig. 13. These two surfaces can be discriminated at scales below about 200 000 μm^2, with no apparent lower limit.

Differences in surface roughness within specimens

The lack of ability to discriminate the relative areas from inside the tool marks to those from the measurements of specimen 2 is shown in Fig. 14. These two surfaces cannot be discriminated at any scales. They appear to be similar. The large mean square ratios at scales above the smooth rough crossover should be dismissed as statistical anomalies and are unimportant because the differences in the relative areas are small.

The ability to discriminate the relative areas from inside the tool marks to those from outside the tool marks on the measurements of specimen 3 is shown

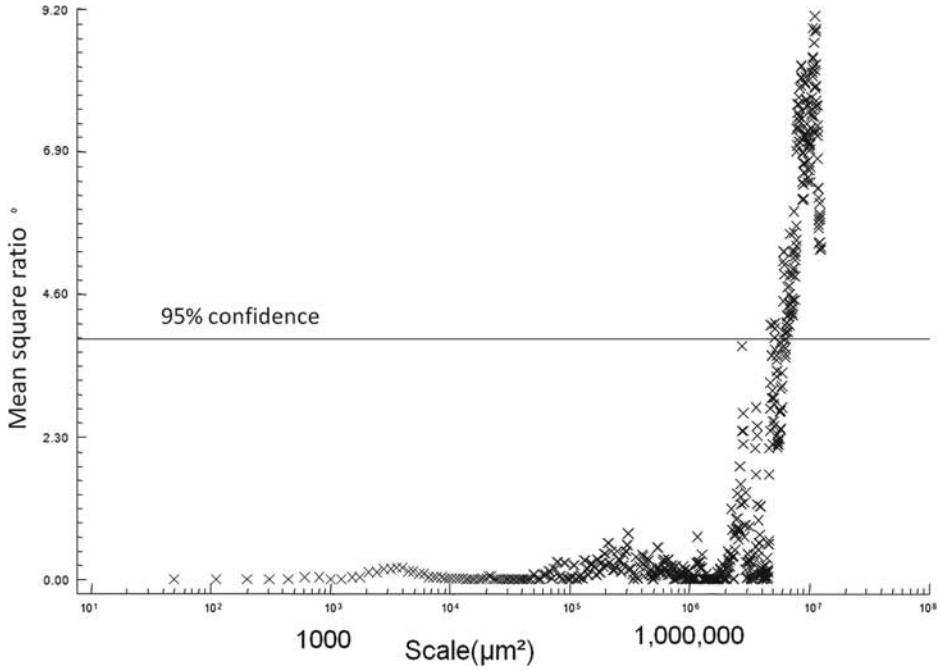

FIG. 14—*Mean square ratios (MSR, which is unitless) versus scale from an F-test comparing inside the tool marks to outside the tool marks on specimen 2; horizontal line is the critical minimum MSR for 95 % confidence that the surfaces are different over the represented scales.*

in Fig. 15. These two surfaces can be discriminated at scales below about 100 000 μm^2, with no apparent lower limit.

Discussion

This work appears to show significant differences in the roughness of the three natural plywood surfaces that have been prepared similarly by the procedures previously described in the method section. The differences are evident from modified F-tests of the relative areas as a function of scale. The relative-areas are calculated using area-scale analysis on topographic measurements from the scanning laser profiler [22]. However, within specimens, the tool mark regions are not consistently different from the surrounding regions outside the tool marks. The inability to discriminate regions inside and outside the tool marks tends to dismiss the hypothesis that the tool has modified the fine scale topography of the wood surface inside the tool mark. At least the topography has not been modified in a way that can be detected by the kind of measurement used in this work when characterized with area-scale analyses. Therefore, if particle adhesion can be linked to the relative areas at the fine scales studied here, as detected by the measurement and analysis methods used here, then there

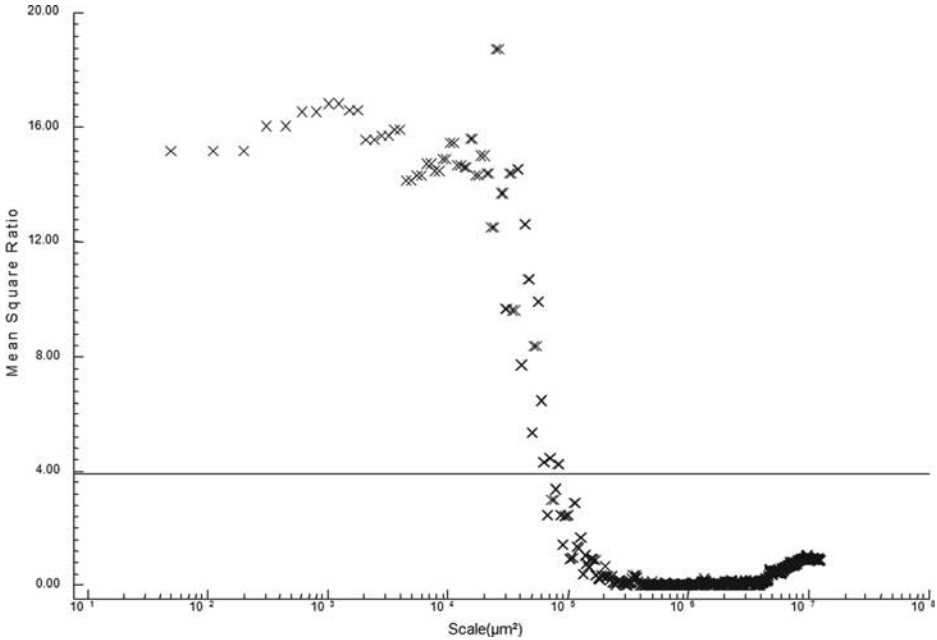

FIG. 15—*Mean square ratios (MSR, which is unitless) versus scale from an F-test comparing inside the tool marks to outside the tool marks on specimen 3; horizontal line is the critical minimum MSR for 95 % confidence that the surfaces are different over the represented scales.*

should not be a difference in the particle adhesion inside the depressions left by the tools compared with the particle adhesion outside the tool marks. If there is any tendency for the tool marks to enhance the retention of particles, then it could be at other scales than those analyzed here.

For example, in specimen 2 there are a series of parallel grooves that might possibly hinder effective cleaning or sampling with a wipe and therefore there could be more particles remaining in the groove than outside the groove. However, the retention of particles due to the topography at finer scales, on the order of 75 μm, will not differ within the groove to outside the groove, provided that the topographic measurement and analysis methods used here are indicative of particle adhesion.

On surface 3 the relative areas at areal scales below about 60 000 μm^2 can consistently be used to discriminate the regions inside the tool mark from outside the tool marks (Fig. 14). This areal scale corresponds to a linear scale of 346 μm. Supposing that the surface inside the tool marks is smoother than outside, this result suggests that particles with diameters less that 346 μm might have a lesser tendency to be retained in the surface roughness inside the tool mark than outside. If this is true then on surface 3 the relative difficulty of removing particles from within the tool mark versus outside might be compensated somewhat by

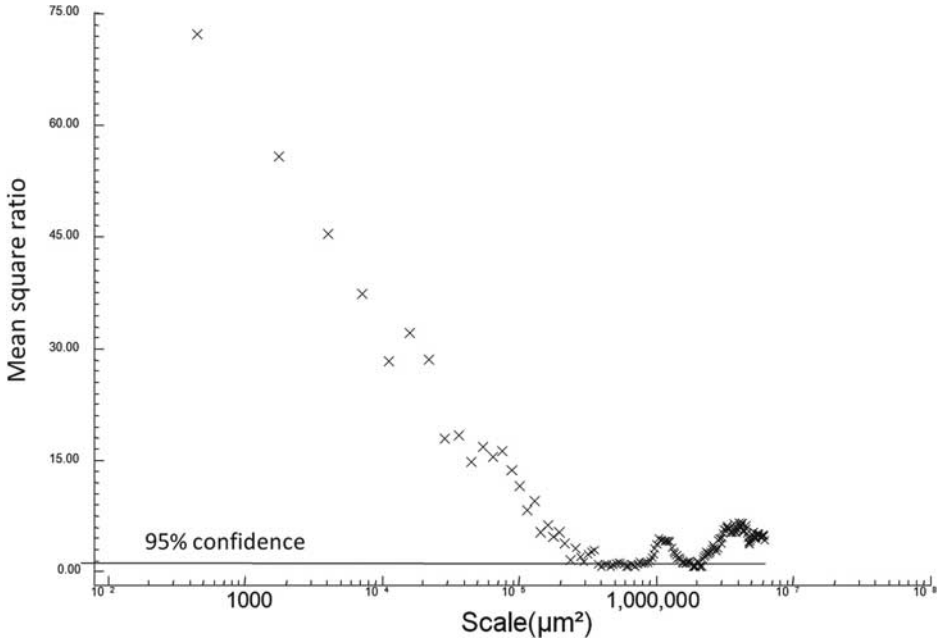

FIG. 16—*Mean square ratios versus (MSR, which is unitless) scale from an F-test comparing coated (latex paint) versus uncoated (polyurethane) finishes; horizontal line is the critical minimum MSR for 95 % confidence that the surfaces are different over the represented scales.*

the relative ease of removing particles from the relatively smoother surface inside the tool mark.

There is much that can still be discovered about the topography of wood surfaces, their modifications by tools, and about the tendency of topography to influence cleaning. The observation that within specimen 2 no difference can be found inside the tool marks versus outside the tool marks while on specimen 3 a difference can be found, is sufficient to conclude that there is no systematic difference in the roughness caused by the tool. There is no general statement that can be made in regard to the tools changing the texture of the wood inside the tool marks, at least detectable by the means used here. Nonetheless, significant differences are found in the surfaces themselves, regardless of the tool marks. The nature of the differences between the surfaces also depends if you are comparing specimen 1 and 2 (Fig. 12) or surfaces 2 and 3 (Fig. 12). The topographies and how they are influenced by the tools differs between specimens 1, 2, and 3.

The largest scales where the wood surfaces show differences are about 30 000 μm^2. The finest scales where the wood surfaces show differences are less than 200 μm^2. A standard reference material, Lead in Soil (3,000 ppm Pb), obtained from the National Institute of Standards and Testing, was deposited on these surfaces in the cleaning studies [15,16] (and the particles were less than 75 μm in diameter, corresponding to an areal scale (area of a circle) of about 4400 μm^2 [26].

The particle size provides an indication of a possible scale of interaction for particle retention and removal. This possible scale of interactions, based on the particle size, is within the scales of discrimination noted above.

Some preliminary surface roughness measurements were taken of the painted surfaces and they were significantly different than the polyurethane coated specimens reported in this paper. A comparison of the relative areas shows that the coating has a profound influence on lowering the roughness of the wood as indicated by relative areas at scales below about 200 000 μm^2 (Fig. 16). The scale of the particle is well within the scales where the coated and uncoated surfaces are clearly different.

This study demonstrated that surface roughness between and within wood specimens that have tool marks can exist but may be at such a fine scale that it is not detected by using cleaning methods. To determine the type of surface roughness that is needed to induce a major influence on removal of leaded dust or other contaminants that deposit on wood surfaces in houses, it would be helpful to develop more profoundly different surface topographies that could be used to evaluate the effectiveness of cleaning as a function of surface. It is also necessary to learn more about the interactions, at various scales, between particle deposition, adhesion, and removal for both cleaning and surface sampling.

Residential and even commercial surfaces rarely feature perfectly smooth, non-porous substances, so studies that make use of a number of surface types can further our knowledge of sampling efficiency [5]. Standards on surface sampling or cleaning such as those described in the introduction, could benefit from further development of the specimens in this study or other coupon size, reproducible specimens. If a collection of standard reference-surfaces were available to researchers and those developing standards, analogous to standard reference substances, then understanding sampling efficiency for differing wipes and other methods would be enhanced.

Conclusions

The method presented here created replicate-stamped wood surfaces that appeared to be representative of the originals. In the future, we would recommend the use of a hydraulic plate press rather than a roller press to reduce warping of samples and a larger sample size to increase the range of surface textures, but this represented a highly successful method for creating reproducible, realistic surfaces for use in cleaning or sampling investigations or standard development.

Specimens did show statistical differences in surface roughness at relatively fine scales of surface roughness. These are scales similar to particle sizes. There is more work that could be done to develop more uniform, standard reference surfaces, with well characterized textures, that can be used to evaluate adhesion and removal of lead, allergens, fungi, endotoxin, bacilli, and chemical contaminants from residential and commercial environments. The surfaces created here are a first step toward development of standard reference material surfaces that could be used to evaluate the performance of sampling or cleaning techniques as standards are devised or revised.

Acknowledgments

The development of the wood specimens was performed for a HUD study entitled, "Evaluation of New Technologies and Methods for Removal of Lead from Hard Surfaces", MOLHT0104-05. The writers acknowledge the assistance of Robert F. Weisberg, Ph.D., for his suggestions in development and conduct of this project. The writers would like to thank Surfract Company for the software used for scale-sensitive fractal analysis, owned by co-author Brown; Solarius Development Corp. for the measurement instrument; and Digital Surf for use of MountainMap software for the thresholding and conventional analysis.

References

[1] Edmonds, J. M., "Efficient Methods for Large-Area Surface Sampling of Sites Contaminated with Pathogenic Microorganisms and Other Hazardous Agents: Current State, Needs, and Perspectives," *Appl. Microbiol. Biotechnol.*, Vol. 84, No. 5, 2009, pp. 811–816.

[2] Lioy, P. J., Freeman, N. C. G., and Millette, J. R., "Dust: A Metric for Use in Residential and Building Exposure Assessment and Source Characterization," *Environ. Health Perspect.*, Vol. 110, No. 10, 2002, pp. 969–983.

[3] ASTM, D7659-10, 2010, "Standard Guide for Strategies for Surface Sampling of Metals and Metalloids for Worker Protection,"ASTM International, West Conshohocken, PA.

[4] ASTM D7144-05a, 2011, "Standard Practice for Collection of Surface Dust by Micro-Vacuum Sampling for Subsequent Metals Determination," ASTM International, West Conshohocken, PA.

[5] Ashley, K., Applegate, G. T., Wise, T. J., Fernback, J. E., and Goldcamp, M. J. "Evaluation of a Standardized Micro-Vacuum Sampling Method for Collection of Surface Dust," *J. Occup. Environ. Hyg.*, Vol. 4, No. 3, 2007, pp. 215–223.

[6] Lewis, R.D., Breysse, P. N., Lees, P. S., Diener-West, M., Hamilton, R. G., and Eggleston, P., "Factors Affecting the Retention of Dust Mite Allergen on Carpet," *Am. Ind. Hyg. Assoc. J.*, Vol. 59, No. 9, 1998, pp. 606–613.

[7] ASTM D4185-06, 2006, "Standard Practice for Measurement of Metals in Workplace Atmospheres by Flame Atomic Absorption Spectrophotometry," ASTM International, West Conshohocken, PA.

[8] ASTM D 6966-08, 2008, "Standard Practice for Collection of Settled Dust Samples Using Wipe Sampling Methods for Subsequent Determination of Metals," ASTM International, West Conshohocken, PA.

[9] ASTM E1613-04, 2004, "Standard Test Method for Determination of Lead by Inductively Coupled Plasma Atomic Emission Spectrometry (ICP-AES), Flame Atomic Absorption Spectrometry (FAAS), or Graphite Furnace Atomic Absorption Spectrometry (GFAAS) Techniques," ASTM International, West Conshohocken, PA.

[10] ASTM E1644-04, 2004, "Standard Practice for Hot Plate Digestion of Dust Wipe Samples for the Determination of Lead," ASTM International, West Conshohocken, PA.

[11] ASTM E1728-10, 2010, "Standard Practice for Collection of Settled Dust Samples Using Wipe Sampling Methods for Subsequent Lead Determination," ASTM International, West Conshohocken, PA.

[12] ASTM E1792-03, 2008, "Standard Specification for Wipe Sampling Materials for Lead in Surface Dust," ASTM International, West Conshohocken, PA.

[13] ASTM E1979-04, 2004, "*Standard Practice for Ultrasonic Extraction of Paint, Dust, Soil, and Air Samples for Subsequent Determination of Lead*," ASTM International, West Conshohocken, PA.

[14] U. S. Department of Housing and Urban Development (HUD), *Guidelines for the Evaluation and Control of Lead-Based Paint Hazards in Housing*, Office of Healthy Homes and Lead Hazard Control, Ed., U.S. Dept. of Housing and Urban Affairs, Washington, D.C., 1995.

[15] Emo, B., Ong, K. H., Kennedy, J., Thummalakunta, L. N. A., and Lewis, R.D., 2007, "Evaluation of New Technologies and Methods for Removal of Lead from Hard Surfaces," in *American Industrial Hygiene Conference*, Philadelphia, PA, June 2-7, 2007, American Industrial Hygiene Association, Fairfax, VA.

[16] Lewis, R. D., Ong, K. H., Kennedy, J., Emo, B. M., Thummalakunta, L. N., and Condoor, S., *Evaluation of New Technologies and Methods for Removal of Lead from Hard Surfaces*, U. S. Dept. of Housing and Urban Development, Office of Healthy Homes and Lead Hazard Control, Ed., U.S. Dept. of Housing and Urban Affairs, Washington, D.C., 2008.

[17] Lewis, R. D., Condoor, S., Batek, J., Ong, K. H., Backer, D., Sterling, D., Siria, J., Chen, J. J., and Ashley, P., "Removal of Lead Contaminated Dusts from Hard Surfaces," *Environ. Sci. Technol.*, Vol. 40, No. 2, 2006, pp. 590–594.

[18] American Society of Mechanical Engineers, *ASME/ANSI B46.1 2002, Surface Texture: Waviness, Roughness, and Lay*. American Society of Mechanical Engineers, New York, 2002.

[19] Jordan, S. E. and Brown, C.A., "Comparing Texture Characterization Parameters on Their Ability to Differentiate Ground Polyethylene Ski Bases," *Wear*, Vol. 261, No. 3–4, 2006, pp. 398–409.

[20] Altin, C., Huynh, T., Powers, B., Brown, C. A., Wang, S., and Zhang, J. S. "Assessing Surface Roughness for Correlation with Particulate Deposition," *IAQVEC (2010), the 7th International Conference on Indoor Air Quality, Ventilation and Energy Conservation in Buildings*, Syracuse, NY, August 15-18, 2010, Department of Mechanical and Aerospace Engineering, Syracuse University.

[21] ASTM, *ASTM D1037-06a Standard Test Methods for Evaluating Properties of Wood-Base Fiber and Particle Panel Materials*, ASTM International, West Conshohocken, PA, 2006.

[22] Brown, C. A., Charles, P. D., Johnsen, W. A., and Chesters, S., "Fractal Analysis of Topographic Data by the Patchwork Method," *Wear*, Vol. 161, No. 1–2, 1993, pp. 61–67.

[23] Berglund, J., Brown, C. A., Rosen, B.-G., and Bay, N., "Steel Surface Roughness Correlation with Steel Sheet Friction," *CIRP Ann. Manuf.*, Vol. 59, 2010, pp. 577–580.

[24] Lipson, C. and Sheth, N. J., *Statistical Design and Analysis of Engineering Experiments*, McGraw-Hill, New York, 1973.

[25] Brown, D. R. S. and Brown, C. A., "Investigation of Surface Topography Differences in Native and Exotic Invertebrates in the St. Lawrence River," *Scanning*, Vol. 32, No. 4, 2010, pp. 250–255.

[26] Montgomery, R. R., and Sauerwein, J. C., *SRM NIST Standard Reference Materials Catalog, NIST SP-1104*, National Institute of Standards and Technology, National United States Department of Commerce, Washington, D.C.

Reprinted from JAI, Vol. 8, No. 10
doi:10.1520/JAI103482
Available online at www.astm.org/JAI

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Assessment of the Bacterial Contamination and Remediation Efficacy After Flooding Using Fluorometric Detection

ABSTRACT: In the aftermath of flooding, rapid response assessment and cleanup is critical to recovery. In this study, the level of contamination of bacteria on surfaces in flooded houses was estimated and compared to the level found on surfaces in non-flooded houses with a rapid field test based on hydrolase enzyme activity present in bacteria and by measuring endotoxin. Sampling was performed by swabbing a 9 cm^2 area using sterile cotton swabs wetted with a bacteriostatic buffer. A correlation between endotoxin levels and levels of hydrolase activity was seen ($R^2 = 0.6469$, $P < 0.0001$). The median value and the variance ofthe result distribution were higher in flooded buildings as compared to non-flooded buildings. In the non-flooded buildings, surfaces were divided into visually clean and visually dirty. As expected the level of bacteria was higher on the visually dirty surfaces, and overall the hydrolase activity correlated well with the visual inspection. Using the results from the visually clean surfaces in the reference buildings as the criteria for clean, four methods of cleaning were tested for their ability to reach these criteria.

KEYWORDS: bacteria, hydrolase activity, flooding, endotoxin, post remediation verification, cleaning

Introduction

Evaluating tools for assessment of bacterial contamination after flooding events or sewage overflow should address whether the measured parameter is relevant,

Manuscript received October 14, 2010; accepted for publication July 15, 2011; published online August 2011.
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Cite as: Reeslev, M., Nielsen, J. C. and Rogers, L., "Assessment of the Bacterial Contamination and Remediation Efficacy After Flooding Using Fluorometric Detection," *J. ASTM Intl.*, Vol. 8, No. 10. doi:10.1520/JAI103482.

reproducible, and rapid. Traditionally, assessment has been performed using cultivation of coliform and fecal indicator bacteria like *Escherichia coli*. Assessments have rightfully focused on the risk of infection as the main health concern. However, health effects of bacterial contamination are not solely a question of risk of infections from pathogenic bacteria. A high number of bacteria in general can cause other health effects such as hypersensitivity pneumonitis, allergy, and inflammation due to the influence of bacterial cell wall agents. One such example is endotoxin, which is part of the outer cell membrane of all Gram-negative bacteria. It can be released in water suspensions after lysis of the bacteria. During agitation of water and dust, it can become aerosolized and increased levels of endotoxin have been measured in many studies and related to health effects [1–4]. Thus measurements of total bacteria level as well as infectious bacteria are relevant for risk assessments.

Another issue with evaluating contamination and remediation cleaning efforts based on cultivation of *E. coli* is that this approach suffers from the fact that *E. coli* and many pathogenic bacteria typically have a very short survival time in the environment. Therefore, noninfectious bacterial reservoirs can remain undetected on furnishings, carpet, and other surfaces in the flooded buildings. Testing for endotoxin has an advantage over cultivation in that endotoxin is relatively stable and will remain detectable for a longer period even when many of the bacteria are dead or unculturable [5].

To quantify endotoxin, it needs to be extracted from the bacterial membrane. This is a relatively complex process, and the outcome is very dependent on the protocol used conferring an inherent variability to endotoxin assays [6,7]. Many protocols exist and with significant differences in yield of endotoxin extracted. Some important variables in the protocols are agitation rate and time, use of ultrasound, and addition of the surface active compound Tween. The lack of standardization makes it difficult to compare endotoxin levels between studies when different protocols are used. Furthermore, at present, endotoxin analysis is a time consuming, costly laboratory-based process, and there is no field detection kit for endotoxin.

In an attempt to improve the assessment of bacteria in environmental samples, this study compares a field detection method for fluorometric detection of hydrolase activity to endotoxin levels. Measuring enzymatic activity is simple and occurs without any extraction procedures. The fluorophore that is released upon hydrolyzation of a fluorogenic enzyme substrate is released extracellularly and can be measured directly. While cultivation of bacteria typically takes several days, hydrolase measurements can be performed on location in less than 1 h.

The present study has been conducted to evaluate if bacterial hydrolase activity could be used to measure of the level of bacterial contamination to evaluate whether the levels of bacterial hydrolase activity were significantly different between visually clean and visually dirty surfaces in reference buildings and between visually dirty surface in reference buildings and contaminated surfaces in flooded buildings. Finally, the study sought to evaluate whether different cleaning efforts in a flooded building could bring the level of bacteria from contaminated to clean where clean would refer to the level as found on visually clean surfaces in reference buildings.

Materials and Methods

Selecting Monitoring Sites

Buildings that had been flooded due to heavy rain and with suspected sewage contamination were termed flooded buildings. Sampling was performed when drying had been completed or was in the late phase.

Buildings with no prior history of flooding were visually inspected for moisture problems or other signs of water damage. If no problems were discovered, the building was used as a non-flooded or reference building.

Sampling

Sterile cotton swabs on wooden handles were moistened with a buffer containing a bacteriostat. Moistened swabs have a greater sampling efficiency [9], and the bacteriostat prevents growth on the cotton swab without affecting the hydrolase activity of the bacteria present. For each sample, an area of 9 cm^2 was swabbed. The swabs were then used to analyze for hydrolase activity and endotoxin.

Enzyme Activity

Bacterial hydrolase activity was measured typically less than 24 h after sampling using the standard operating procedure specified by the manufacturer's specifications (Mycometer A/S, Copenhagen, Denmark). The swabs were transferred to a tube containing 2 ml of the fluorogenic 4-methylumbelliferone hydrolase enzyme substrate and allowed to react for approximately 30 min. The exact reaction time is dependent on the ambient temperature and was determined according to a table listed in the manufacturer's manual. The fluorescence generated during the reaction time was measured by withdrawing 100 μl of the reactant and adding it to 2 ml of alkaline buffer in a disposable polystyrene cuvette (Sarstedt AG, Nümbrecht, Germany). The fluorescence was measured on a Picofluor fluorometer (Turner Designs, Sunnyvale, CA) using an excitation wavelength of 365 nm and measuring emission at 445 nm. Calibration of the instrument was performed prior to analysis using two standards of known fluorescence. The calibration was acceptable if the instrument reading of the fluorescence standard deviated less than 1% from the fluorescence value of the standard. Enzyme substrate blanks were measured and subtracted from sample values. Hydrolase activity is measured in arbitrary fluorescence units (AFU). Analytical grade chemicals and pyrogenic free water was used for preparing the buffers.

Endotoxin

Samples for endotoxin analysis were frozen at − 50°C until all samples had been collected. Endotoxin was extracted from the swabs by agitating for 15 min in pyrogenic free water. The extracts were diluted 10^3 to 10^6 times in pyrogenic

free water before analysis. Five sterile swabs were extracted by the same procedure and the mean value used as the blank value.

The amount of endotoxin in the liquid extracts was determined using the Limulus Aemobocyte Lysate method with specific lysate and the chromogenic, kinetic version (Associates of Cape Cod, MA). The 405 nm absorbance of the samples was measured during a 30 min reaction time on a SK 601 Wellreader (Seikagaku Corporation, Tokyo, Japan). The incubation temperature was 37°C. Four different endotoxin concentrations were prepared according to the manufacturer's specifications and used as internal standards. All glassware was baked at 270°C for 3 h before use. The results are given in nanograms endotoxin.

Results and Discussion

Bacterial Hydrolase Activity and Heterotrophic Plate Count (HPC)

A small amount of yeast extract (125 mg/l) was added to tap water to simulate an organic pollution. The indigenous bacteria in the tap water were allowed to proliferate reaching a maximum after approximately 30 h. A dilution series was prepared, and samples were withdrawn for HPC and hydrolase activity. Figure 1 shows that the activity of hydrolase activity correlates to the level of bacteria determined by HPC in water samples with different levels of indigenous bacteria ($R^2 = 0.988$).

Bacterial Hydrolase Activity and Endotoxin

Parallel samples were collected in both flooded and non-flooded reference buildings and used to analyze for hydrolase activity and endotoxin. Figure 2 shows a

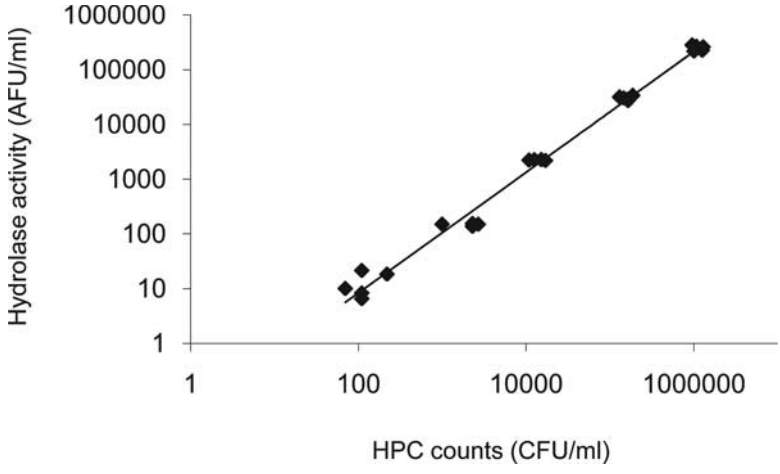

FIG. 1—*Correlation between bacterial hydrolase and HPC counts ($R^2 = 0.988$).*

correlation between the two bacteria indicators ($R^2 = 0.6469$, $P < 0.0001$). The two parameters correlate despite that endotoxin is only present in gram negative bacteria while the hydrolase activity is present in both gram negative and gram positive bacteria (unpublished result).

The endotoxin levels measured in this study varied from below the detection limit to a maximum level around 2800 ng/cm^2. Surprisingly studies describing the level of endotoxin on surfaces in buildings were not found in the literature search. The quite extensive research on endotoxin in the environment focuses mainly on air measurements and to some extent dust samples. The levels found in the present study are quite high, e.g., compared to the guidelines proposed for "no-effect level" for environmental endotoxin [8]. This guideline describes health effects of increasing seriousness with endotoxin concentrations ranging from 10 to 200 ng/m^3. With a maximum concentration of 2.8×10^7 m^2 found in the present study and assuming an average ceiling height of 2.5 m, 1 m^2 of the floor surface feeds approximately 2.5 m^3 of air above. This means that less than one thousands of a percentage of the endotoxin on the surface would have to become airborne to exceed the high level (200 ng/m^3).

Flooded Versus Reference Buildings and Categories

Swab samples were taken from surfaces in buildings with flooding or sewage spills. For comparison, samples were also taken from reference buildings with no flooding or sewage spills. Samples from the reference buildings were divided into samples from visually clean surfaces and from surfaces with varying levels of visual dirt. The results from the visually clean surfaces were used as criteria for clean after remediation. The results from the visually dirty surfaces in the reference building were used to describe dirty but not contaminated surfaces. For a surface to be classified as contaminated in the flooded buildings, the level

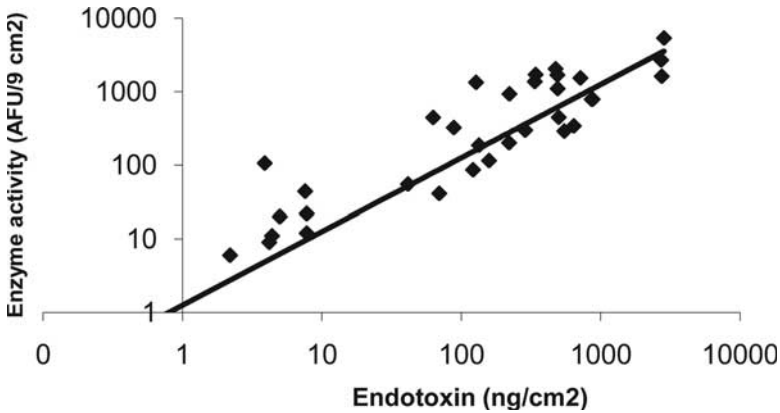

FIG. 2—*Correlation between the level of bacterial hydrolase activity and endotoxin in samples from surfaces of flooded and reference buildings ($R^2 = 0.6469$, $P < 0.0001$).*

TABLE 1—*The minimum, maximum, mean, standard deviation, standard error of the mean 95% confidence intervals, and median values of hydrolase activity measured on swab samples from surfaces of reference buildings and flooded buildings. The values given is a measure of the hydrolase activity expressed in arbitrary fluorescence units (AFU) according to the manufacturer's protocol.*

	Number of Samples	Median Value, AFU	Mean Value, AFU	STD, AFU	SEM, AFU	95% CI, AFU	Minimum and Maximum Values, AFU
Reference buildings Visual clean surfaces	88	5	5	4	0.4	(4.2;5.8)	0: 20
Reference buildings Visual dirty surfaces	110	15	50	79	7.5	(35;65)	0: 392
Flooded buildings	169	226	734	1866	144	(446;1022)	3: 15082

of hydrolase activity should be above the level found on the dirty surfaces in the non-flooded buildings.

Table 1 shows a marked difference in the hydrolase activity from the different categories of surfaces. A logarithmic normal distribution was seen with samples from both flooded and non-flooded buildings. The visual clean surfaces in the reference buildings had a median value of 5 AFU, which is also the detection limit of the method, and all values were below 20 AFU. The median value, mean value and the variance were higher on visually dirty surfaces in non flooded buildings and much higher in flooded buildings.

Measuring Cleaning Efficacy

Measurements were made in a living room of a house that had been flooded after heavy rain. The floor had been removed, and the room was dried out. Four areas (1 m^2 each) of the concrete foundation were cleaned using four different cleaning methods. All areas were vacuumed with a vacuum cleaner with HEPA filter. Three of the parcels were then cleaned thoroughly with one

TABLE 2—*Level of bacteria measured as hydrolase activity on surfaces in a flooded house before and after cleaning. Four different cleaning treatments were used. Cleaning using steam and three types of chemistry were tested. Sampling was in duplicate.*

Treatment	n	Before cleaning, AFU Mean (STD)	n	After cleaning, AFU Mean (STD)
Steam cleaning	2	434(26)	2	7(0.0)
Rodalon (quaternary ammonium)	2	408(88)	2	2.0(1.4)
Biowash (quaternary ammonium)	2	419(76)	2	10.5(2.1)
Peroxy tabs (peroxide)	2	875(222)	2	44.5(4.9)

of three cleaning chemicals. Rodalon, (a quaternary ammonium chloride), Biowash (a quaternary ammonium chloride), and peroxide tabs (potassium persulfate, oxidizing agent). The fourth parcel was cleaned with a steam method. Steam, at 6 bars with temperature from 130 to 150°C, was blown onto the surface. This procedure was repeated a second time with the steam passing through a microfiber cloth that is in contact with the surfaces and absorbs released dirt/bacteria. Finally, all four areas were vacuumed again before testing for enzyme activity. The results are shown in Table 2. All treatments significantly reduced the bacterial level. Three of four treatments were able to reduce the level of bacteria to a level not distinguishable from visually clean surfaces in the reference buildings.

References

[1] Rylander, R., "Endotoxin in the Air—Good or Bad for You?" *Clin. Pulm. Med.*, Vol. 14, 2007, pp. 140–147.

[2] Kennedy, S. M., Christiani, D. C., and Eisen, E. A., "Cotton Dust and Endotoxin Exposure-Response Relationships in Cotton Textile Workers," *Am. Rev. Respir. Dis.*, Vol. 135, 1987, pp. 194–200.

[3] Smid, D., Heederik, D., Houba, R., and Quanjer, P. H., "Dust and Endotoxin Related Respiratory Effects in the Animal Feed Industry," *Am. Rev. Respir. Dis.*, Vol. 146, 1992, 1474–1479.

[4] Vogelzang, P. F., van der Gulden, J. W., Folgering, H., Kolk, J. J., Heederik, D., Preller, L., Tielen, M. J., and van Schayck, C. P., "Endotoxin Exposure as a Major Determinant of Lung Function Decline in Pig Farmers," *Am.J. Respir. Crit. Care Med.*, Vol. 157, 1998, pp. 8–15.

[5] Dingman, J. R., Rayner, M. G., Mishra, S., Zhang, Y., Ehrlich, M. D., Post, J. C., and Ehrlich, G. D., "Correlation Between Presence of Viable Bacteria and Presence of Endotoxin in Middle Ear Effusions," *J. Clin. Microbiol.*, Vol. 36, No. 11, 1998, pp. 3417–3419.

[6] Morris, N. M., Catalano, E. A., and Berni, R. J., "3-Hydroxymyristic acid as a measure of endotoxin in cotton lint and dust," *Am. Ind. Hyg. Assoc. J.*, Vol. 49, No. 2, 1988, pp. 81–88.

[7] Milton, D. K., Gould, M. C., Roslansky, P. F., and Novitsky, T. J., "New Estimates of Endotoxin Content of cotton Dust: SDS Suspensions and Kinetic LAL Test," *Proceedings of 12th Cotton Dust Research Conference*, R. R. Jacobs and P. J. Wakelyn, Eds., National Cotton Council, Memphis, TN 1987, p. 197.

[8] Rylander, R., Ed., "Endotoxins in the Environment—A Criteria Document," Int. J. Occup. Environ. Health, Vol. 3, 1997, pp. S1–S48.

[9] Rose, R., Jensen, B., Peterson, A., Banerjee, S. N., and Arduino, M. J., "Swab Materials and Bacillus anthracis Spore Recovery From Nonporous Surfaces," *Emerg. Infect. Dis.*, Vol. 10, No. 6, 2004, pp. 1023–1029.

Reprinted from JAI, Vol. 8, No. 6
doi:10.1520/JAI103481
Available online at www.astm.org/JAI

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Use of Direct Reading Surface Sampling Methods for Site Characterization and Remediation of Methamphetamine Contaminated Properties†

ABSTRACT: Residual methamphetamine contamination in Clandestine laboratories represents a hazard to emergency response personnel, remediation workers and the general public. To address this threat, two rapid, sensitive surface sampling techniques to assess the location and level of methamphetamine contamination were developed. Both methods employ established industrial hygiene surface sampling materials (wipes and swabs) but differ in their sensitivity and detection technology. One method, based on colorimetric disclosure, detects and confirms a collected sample or visible residues. The second method uses a lateral flow immunochemical assay

Manuscript received October 14, 2010; accepted for publication April 21, 2011; published online July 2011.

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Presented at the ASTM International Symposium on Surface and Dermal Sampling, 14–15 October 2010, San Antonio, TX.

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Cite as: Snawder, J. E., Striley, C. A. F., Esswein, E. J., Hessel, J., Sammons, D. L., Robertson, S. A., Johnson, B. C., MacKenzie, B. A., Smith, J. P. and Walker, C. V., "Use of Direct Reading Surface Sampling Methods for Site Characterization and Remediation of Methamphetamine Contaminated Properties," *J. ASTM Intl.*, Vol. 8, No. 6. doi:10.1520/JAI103481.

(LFIA) for semi-quantitative detection of trace contamination. The National Institute for Occupational Safety and Health (NIOSH) partnered with public health agencies to develop applications of the methods for assessment of methamphetamine contamination of suspected properties. These applications focused on safe strategies for site assessment, hazard characterization, and remediation effectiveness. To conduct the field studies, NIOSH researchers and their partners visited more than a dozen suspected laboratories including mobile labs, abandoned properties, occupied residences, and motel rooms. NIOSH found greater than 95% agreement between positive identification of the presence of methamphetamine by LFIA and laboratory-based, liquid chromatography mass spectroscopy (LC–MS) methods. Test results were used to develop site assessments and make personal protective equipment recommendations. Results were also used to conduct process-based decontamination of properties and to make health-based decisions on remediation, re-occupancy of residences, as well as determine the degree of contamination of personal property in an inactive clandestine laboratory. By partnering with stakeholders, NIOSH was able to achieve two primary goals: (1) to develop a level of awareness in health department sanitarians, law enforcement personnel and other first responders that methamphetamine surface contamination was a potentially significant route of exposure; (2) to validate our methods in the field and to develop protocols for proper use and interpretation of the results.

KEYWORDS: clandestine lab, methamphetamine, surface wipe, real-time, direct reading

Introduction

According to the United States Drug Enforcement Agency (US DEA) discovery of clandestine methamphetamine laboratories peaked at 17 000 in 2003–2004. State and federal laws restricting availability of methamphetamine precursors, particularly pseudoephedrine or ephedrine, have led to initial decreases in Clandestine laboratory discoveries or seizures [1]. However, thousands are still found each year (http://www.justice.gov/dea/concern/map_lab_seizures.html). Small-scale methamphetamine laboratories supply approximately 20% of the US methamphetamine supply [2,3], and this number is expected to increase [4]. Residual contamination of clandestine methamphetamine laboratories represents a hazard to emergency response personnel, remediation workers and the general public [5–7].

In a series of studies, researchers from National Jewish Health and the National Institute for Occupational Safety and Health (NIOSH) along with law enforcement and public health agencies examined suspect clandestine laboratories for the presence of hazardous chemicals and methamphetamine contamination. These studies looked at exposures after police seizures and during controlled methamphetamine synthesis or "cooks" [8,9] during a control "cook" and the following 24 h [9], and during simulated methamphetamine smoking

[10]. Findings from these studies found that during active methamphetamine manufacture or smoking, airborne exposures to toxic chemicals and methamphetamine were often elevated but airborne concentrations decreased significantly after 24 h. Surface contamination by methamphetamine does remain a significant risk for dermal exposures and transfer from surfaces to unprotected individuals can occur long after manufacturing ceases and is possible at locations far from the actual manufacturing process [8,11].

Reducing risks for methamphetamine exposures involves awareness of surface contamination; especially the risks for contact transfer of methamphetamine to hands and other skin surfaces as the primary route. Permanent cleanup (or remediation) of former clandestine laboratory sites to eliminate hazards posed by residual methamphetamine presents many issues regarding the costs and responsibility for cleanup. State and Federal governments that establish standards for acceptable post-remediation contamination levels often assign responsibility for enforcement of standards to environmental and public health agencies. Because of the high cost of complete remediation, owners often abandon properties rather than undertake the financial burdens of remediation. NIOSH has developed numerous methods for surface sampling and analysis to detect methamphetamine on surfaces.

Three traditional industrial hygiene methods were developed and validated for the NIOSH manual of analytical methods (NMAM), specifically NMAM 9106, 9109, and 9111. The methods all use mass spectroscopy and isotopic dilution but differ in sample preparation and analysis. NMAM 9106 and 9109 are gas chromatography/mass spectroscopy (GC/MS) methods, and 9111 is liquid chromatography/mass spectroscopy (LC/MS) method. NIOSH has also developed a surface plasmon resonance method for real time quantitative analysis of methamphetamine on surfaces [12]. While laboratory methods are sensitive and accurate they do have shortcomings. Surface samples need to be collected, transported to the laboratory and analyzed, a time consuming process requiring specialized equipment and trained personnel. In light of this, NIOSH was contacted by law enforcement and public health agencies to develop rapid tests that could be used in the field with minimal training. This manuscript describes the development and validation of rapid, sensitive surface sampling technologies to assess the location and level of methamphetamine contamination in clandestine labs. Two technologies were developed in tandem. The first method is for initial confirmation of the presence of methamphetamine in bulk samples and surface residues (colorimetric method) and the second, a sensitive, semi-quantitative detection method is used to determine the extent of contamination and assess remediation effectiveness (immunochemical method).

The goal of this study was to validate both the colorimetric and immunochemical surface sampling and detection techniques for methamphetamine by subjecting the methods to laboratory and field validation tests. Accuracy and sensitivity was determined in the laboratory and in field testing. Aside from using the technologies to assure compliance to state and local surface limits, NIOSH partnered with the Hamilton County Health District (OH) to use the tests to: (1) perform risk assessments and assess the potential of cross contamination to workers involved in the demolition of a former laboratory; (2)

evaluated application of the tests to assess decontamination; (3) assess cross contamination from surfaces in rental property that was a former methamphetamine laboratory; and (4) assess the potential for contamination to personal items brought into a contaminated residence.

Experimental

Reagents

All chemicals used in this work were reagent grade or greater purity. Methamphetamine- HCl, acetaldehyde, sodium nitroprusside, sodium carbonate, methanol, phosphate buffered saline, and Triton-X100 were obtained from Sigma–Aldrich (Milwaukee, WI).

Antibodies to methamphetamine and methamphetamine conjugates were purchased from Arista Biologicals Inc. (Allentown, PA). Lateral flow immunoassays (LFIA) were assembled under contract by Arista Biologicals to NIOSH specification.

Deionized water (18 MΩ) was produced using a Barnstead nanopure system (Thermolyne, Dubuque, IA).

Materials

Cotton-tipped wooden swabs (Fisher # 23-400-100), 2×2 gauze wipes (North Safety Products, #041975D) 3×3 gauze wipes (North Safety Products # 041980D), 5 and 10 ml disposable syringes, disposable weigh boats, 50 ml plastic centrifuge tubes, and disposable nitrile laboratory gloves were purchased from Fisher Scientific (Pittsburg, PA). Plastic pump spray bottles (vol. ≈150 ml) were purchased from U.S. Plastics (Lima, OH). Ceramic tiles (U.S. Ceramic Tile 4-1/4 × 4-1/4 in.) were purchased at a local building supply store (Home Depot, Cincinnati, OH).

Sample Handling

All laboratory and field sample collection was carried out while wearing clean nitrile gloves; to prevent cross contamination gloves were changed each time a new test was performed. Appropriate personal protection equipment was worn at all times during laboratory and field procedures. Methamphetamine was stored and standard solutions were prepared in a USDEA licensed laboratory and all research activities were in compliance with USDEA guidelines and rules.

The colorimetric indicator solution was Simon's reagent [13] prepared in a two part solution. Each solution was stored in separate 150 ml plastic spray bottles.

Solution A: 3 g of sodium nitroprusside was dissolved in 150 ml of distilled water and 6 ml of acetaldehyde was added to the solution with thorough mixing.

Solution B: 2% sodium carbonate in distilled water.

Phosphate buffered saline (PBS) containing 0.1% Triton X-100 was used as the immunochemical assay buffer. Dry pre-packaged PBS was dissolved in 1 l of

deionized water and 1.0 ml Triton X-100 was added. Buffer was stored at ambient temperature for up to 14 days.

Methamphetamine stock solutions (1 mg/ml or 100 μg/ml) were prepared in methanol and diluted in PBS-Triton X-100 (0.1%) to the appropriate concentration such that 1 ml applied to the test surface gave the desired concentration.

Procedures

Colorimetric Methamphetamine Test: Laboratory Evaluation and Validation—The colorimetric method allows for rapid sampling, detection and confirmation of methamphetamine in visible residues and suspect surfaces. A surface suspected of being contaminated with methamphetamine is wiped with a cotton pad wetted with PBS-Triton X-100. The presence of methamphetamine is then disclosed on the wipe by applying two sprays of chemical reaction solution (Solution A), followed by four sprays of disclosing solution (Solution B). The results of the test are immediate: a color change to blue indicates the presence of methamphetamine.

For the determination of the linear response of the colorimetric method, serial twofold dilutions (1000–0 μg) of methamphetamine in 100 μl were added to separate wells of 96 well microtiter plates. Solution A (50 μl) and Solution B (100 μl) was added and the plate was transferred to a Molecular Devices SpectraMax plate reader, mixed by shaking and absorbance determined at 550 nm. The procedure was repeated with methamphetamine samples of different purity.

For the determination of sensitivity of the colorimetric method, methamphetamine in sampling buffer was applied to 4×4 in glazed ceramic tiles and allowed to dry undisturbed overnight. Applied methamphetamine ranged from 500 to 0 μg per tile. Tiles were randomized and the test operator was blinded to the level of methamphetamine on each tile. The test operator placed a 10×10 cm template on the area to be sampled, folded the 2×2 in cotton gauze wipe twice to form a sharp edge and wiped the surface to be sampled with firm pressure, using 3–4 vertical S-strokes, followed by 3–4 horizontal S-strokes, and finally wiped the area with 3–4 vertical S-strokes. The sample wipe was placed in a plastic weigh boat, with the portion of the wipe in contact with the sampled surface facing upward. Two sprays of Solution A and four sprays of Solution B were applied to the wipe and observed for formation of a blue color bloom indicative of the positive presence of methamphetamine. Tests were repeated three times with three different operators. Method sensitivity was calculated by plotting test results as a four parameter curve and determining the limit of identification based on the correct identification of the presence of methamphetamine 95% of the time.

Immunochemical Detection of Methamphetamine-Laboratory Validation—An immuno-chromatographic LFIA for the specific detection of methamphetamine was developed. The handheld test gives binary results indicated by the presence of a single line for a positive test of methamphetamine; a negative

test resulted in the formation of two lines. Because different localities and states have different limits for surface residues, test procedures were developed for 50, 100, and 500 ng. To conduct a test the test operator placed a 10×10 cm template on the area to be sampled. The surface to be tested is wiped with a swab or gauze pad moistened with PBS-Triton X-100 as described for the colorimetric method. For swab samples (50 ng), after wiping the surface the swab was placed in a vial containing an extraction solution (PBS-Triton-X-100). After gently shaking the vial, three drops of the solution are dropped on the sample well of the LFIA and the test placed on a level surface to develop. LFIA tests for 100 and 500 ng use cotton gauze wipes and a simple extraction procedure where the sample wipe is placed in a syringe and extraction buffer added. The extracted sample is expressed from the syringe by the plunger and the extract tested as described above.

Determination of Immunochemical Method Accuracy—LFIA accuracy tests were conducted with 10 untrained volunteers. Ceramic tiles were spiked with known concentrations of methamphetamine in methanol and allowed to dry overnight. Following a short training session, volunteers performed wipe tests on individual tiles with either cotton swabs (LFIA 50 ng/ 100 cm^2), or 2×2-inch cotton wipes (LFIA 100ng/ 100 cm^2), or 3×3-inch cotton wipes (LFIA 500 ng/ 100 cm^2) as described above ($n = 480$ tests). Test operators changed gloves between each wipe test to avoid cross contamination. Bayesian analysis was performed on results obtained by volunteers with the following terms and calculations:

TP = true-positive diagnostic test result.

TN = true-negative diagnostic test result.

FN = false-negative diagnostic test result.

FP = false-positive diagnostic test result.

Diagnostic Sensitivity = $[TP/(TP + FN)] \times 100$. Defined as the percentage of positive methamphetamine tests on surfaces with known contamination (spiked laboratory surface).

Diagnostic Specificity = $[TN/(FP + TN)] \times 100$. Defined as the percentage of negative methamphetamine tests on surfaces with no known contamination (clean laboratory surface).

Receiver operating characteristics (ROC) curves were used to compare the analytical sensitivity and specificity of each assay by demonstrating the ability of each test to discriminate between alternative outcome states. Curves were prepared and analyzed using GRAPHROC for Windows (version 2.0; downloaded from http://members.tripod.com/refstat/grdownload.htm). On the *y* axis, sensitivity, or the true-positive fraction, was plotted. On the *x* axis, the false-positive fraction (or 1- specificity) was plotted. The closer the ROC plot is to the upper left corner, the higher the overall accuracy of the test.

Comparison of LFIA Surface Detection Method to NMAM 9111, LC-MS With Isotopic Dilution—Ceramic tiles were spiked with known concentrations of methamphetamine in methanol and allowed to dry overnight. For the LFIA

tests, volunteers performed wipe tests on the tiles with either cotton swabs (LFIA 50), 2×2 cotton wipes (LFIA 100) or 3×3 cotton wipes (LFIA 500) as described above. For detection of methamphetamine on spiked surfaces by liquid chromatography mass spectroscopy (LC-MS) (NMAM 9111), ceramic tiles were spiked with known concentrations of methamphetamine in methanol and allowed to dry overnight. In a separate, methamphetamine-free laboratory, 3×3 wipes were wet with 1 ml reagent grade methanol and individually placed into plastic centrifuge tubes and sealed. Upon entering the detection area, the gauze was taken out of the tubes prior to wiping the designated surface. After sampling, the wipes were put back into the centrifuge tubes and NMAM method 9111 was used for quantitative analysis (ALS Laboratory Group formerly DataChem Laboratories, Salt Lake City, UT).

Field Evaluation and Validation of Tests—The surface wipe methods were field tested in clandestine methamphetamine laboratories in Ohio and Kentucky. Upon invitation from law enforcement and public health agencies NIOSH researchers performed entry into suspect clandestine laboratories. Wipe samples for methamphetamine were collected by wiping a 100 cm^2 area with either sterile cotton swabs for immunochemical analysis, or sterile 3 in. by 3 in. ($3'' \times 3''$) gauze wipes for either colorimetric detection or chemical analysis. Cross contamination of sampling sites was minimized by using separate pairs of gloves between sample locations. On-site methamphetamine detection was performed as described above. For samples that were analyzed by LC/MS, prior to entering the methamphetamine cook area, the 3×3 wipes were wetted with 1 ml of reagent grade methanol and individually placed into plastic centrifuge tubes. After entering the cook area, the gauze was taken out of the tubes prior to wiping the designated surface. After sampling, the wipes were put back into the centrifuge tubes and quantitative laboratory testing by NMAM 9111 was performed (ALS Laboratory Group, formerly DataChem Laboratory Salt Lake City, UT.)

Process-Based Assessment of Decontamination of a Former Methamphetamine Laboratory in a Hotel Room—NIOSH and Hamilton County (OH) Public Health, Environmental Health Division researchers performed initial wipe sampling of a hotel room methamphetamine laboratory immediately after it was seized by law enforcement. Colorimetric, immunochemical and LC/MS (NMAM 9111) samples were taken for an initial assessment of contamination and results were used to advise a commercial cleaning company how to proceed. After initial cleaning efforts, the sample locations were re-tested and the cleaning crew was advised on areas in need of further decontamination. The process of cleaning/re-testing was repeated until all contamination was below method limits.

Assessment of Methamphetamine Transfer From a Contaminated Former Methamphetamine Laboratory to Personal Property Brought into the Residence—The Hamilton County (OH) Public Health, Environmental Health Division was notified regarding health complaints from tenants of a rental property

that was a suspected former methamphetamine laboratory. Law enforcement confirmed that the property had been under surveillance but, no arrests were made. Interviews by personnel from the Environmental Health Division with the property owner confirmed that he had removed materials consistent with a methamphetamine laboratory (pseudoephedrine packaging, solvent containers, etc.,) while cleaning the house between tenants. Based on these findings NIOSH was contacted to assist in sampling the residence and personal property. Over 200 wipe samples were taken for analysis by colorimetric, immunochemical and LC/MS methods. Samples were divided between the structure of the residence and materials brought into the residence by the new tenants. Locations of objects were mapped in relation to permanent structures of the house as well as location in relation to HVAC outlets to determine sources of contamination. Finally, the direct read methods were used by the Environmental Health Division to oversee a process-based decontamination of the property.

Results and Discussion

Colorimetric Methamphetamine Test: Laboratory Evaluation and Validation

The colorimetric method uses Simon's reagent, a solution of sodium nitroprusside and acetaldehyde that reacts with secondary amines to produce a deep blue color under basic conditions. The reagent has been used to identify methamphetamine in urine samples [14] and in bulk samples [13] and commercial tests are available to detect methamphetamine on surfaces. However, until this study

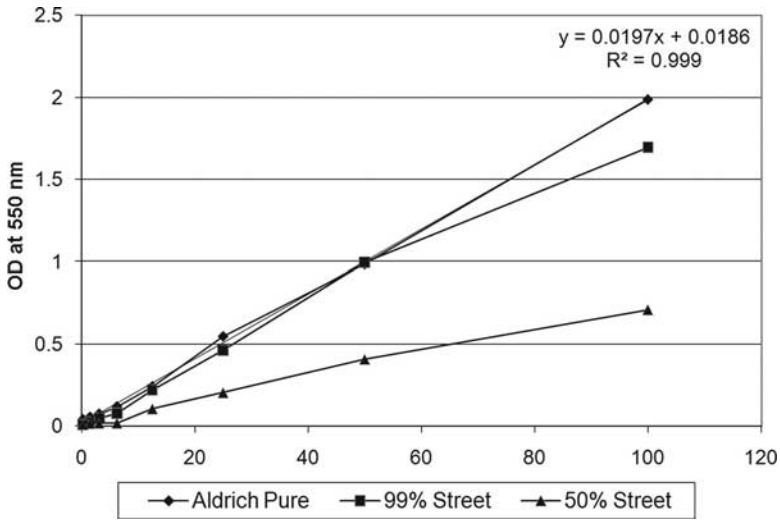

FIG. 1—*Determination of linearity of blue color formation by the reaction of Simon's reagent with methamphetamine. Standard curve of blue color formed (absorbance at 550 nm) by the reaction of methamphetamine (0–100 μg) with Simon's reagent. The procedure was repeated with methamphetamine samples of different purity.*

there had not been a formal validation of the method to determine test accuracy and detection limits on surfaces.

Blue color formed by the reaction of Simon's reagent with methamphetamine was found to be linear from 0 to 500 μg. The intensity of color was directly proportional to the concentration of methamphetamine present. Intensity of color is directly related to the purity of methamphetamine from different sources (Fig. 1).

Based on tests with multiple users and various concentrations of methamphetamine, the colorimetric wipe method limit of identification (LOI) was determined to be 17.3 ± 2.2 μg/100 cm^2 for 95% of users when methamphetamine was present +/-25% stated cut-off (Fig. 2).

Immunochemical Detection of Methamphetamine-Laboratory Validation

The immunochemical sampling and detection methods were found to be accurate and sensitive when used by volunteers with limited training (Tables 1–2). Diagnostic sensitivity was 92% when methamphetamine is present +/−25% stated cut-off $[(259/259 - 21) \times 100]$.

Diagnostic specificity was found to be 100% $[(18/0 - 18) \times 100]$. Method accuracy was greater than 95% to identify presence/absence of methamphetamine

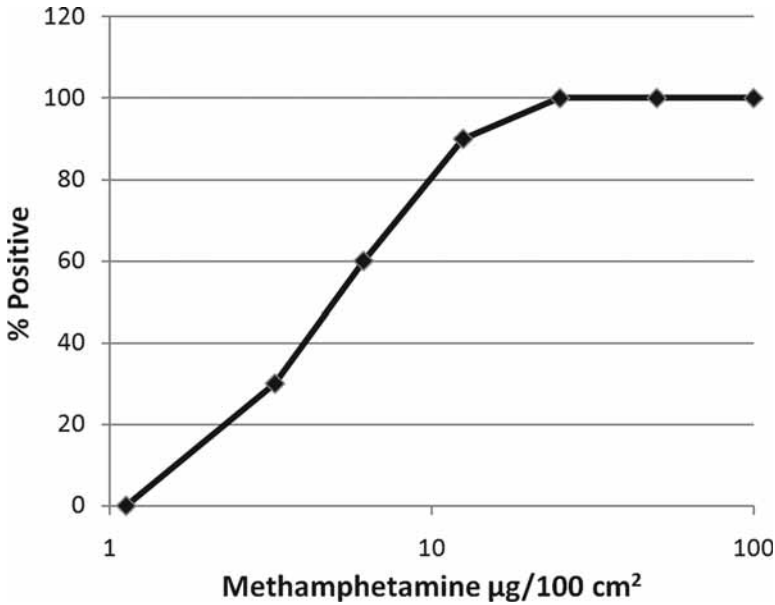

FIG. 2—*Estimation of the colorimetric methamphetamine surface sampling method. Method sensitivity was calculated by plotting test results from three volunteers as a four parameter logistic curve* $[y = y_0 + (a/(1 + ((x/x_o)\text{^}b)))]$. *Method limit of identification (LOI) was determined to be 17.362.2 µg/100 cm^2 for 95% of users when methamphetamine present +/-25% stated cut-off.*

(460/480 correct). Method sensitivity was greater than 95% when methamphetamine was present +/−25% of the stated cut-off. ROC curve analysis found the methods to be very accurate (Fig. 3). Accuracy is measured by calculating the area under the ROC curve (AUC). The LFIA 50, 100, and 500 were all found to have AUCs greater than 0.98. An AUC of 1 represents an ideal test; values near 0.5 represent an indiscriminant test.

Comparison of LFIA Surface Detection Method to NMAM 9111, LC-MS With Isotopic Dilution

In laboratory tests LFIA surface sampling methods were found to have equivalent or greater sensitivity to detect methamphetamine on spiked ceramic tiles compared to NMAM 9111 (Table 3).

TABLE 1—*Determination of the method accuracy of LFIA surface wipe methods for methamphetamine: Method accuracy tests were conducted with 10 untrained volunteers (3 trials/concentration). For the LFIA tests, volunteers performed wipe tests on spiked tiles with either cotton swabs (LFIA 50), 2 × 2 cotton wipes (LFIA 100) or 3 × 3 cotton wipes (LFIA 500) as described in the text (n = 540 tests).*

LFIA 50 [1 ml, 50 ng cutoff (C.O.)]				
Test #	Methamphetamine ng/100 cm^2	% C.O.	Positives(%)	Negatives (%)
1	0	0	0	100
2	50	CO	90	10
3	50	CO	90	10
4	50	CO	100	0
5	40	80	90	10
6	60	120	100	0
LFIA 100 [2 ml, 100 ng cutoff (C.O.)]				
Test #	Methamphetamine ng/100 cm^2	% C.O.	Positives(%)	Negatives (%)
1	0	0	0	100
2	100	CO	100	0
3	100	CO	100	0
4	100	CO	100	0
5	80	80	100	0
6	120	120	100	0
LFIA 500 [10 ml, 500 ng cutoff (C.O.)]				
Test #	Methamphetamine ng/100 cm^2	% C.O.	Positives(%)	Negatives (%)
1	0	0	0	100
2	500	CO	100	0
3	500	CO	100	0
4	500	CO	100	10
5	400	80	90	10
6	600	120	100	0

TABLE 2—*Determination of the method sensitivity of LFIA surface wipe methods for methamphetamine: Method sensitivity tests were conducted with 10 untrained volunteers (3 trials/concentration). For the LFIA tests, volunteers performed wipe tests on spiked tiles with either cotton swabs (LFIA 50), 2 × 2 cotton wipes (LFIA 100) or 3 × 3 cotton wipes (LFIA 500) as described in the text (n = 540 tests).*

LFIA 50 [1 ml, 50 ng cutoff (C.O.)]

Methamphetamine ng/100 cm^2	% C.O.	Positives (%)	Negatives (%)	Equivocal (%)
0	0	0	100	0
25	−50	60	40	0
38	−25	80	20	0
50	C.O.	100	0	0
63	+25	100	0	0
75	+50	100	0	0

LFIA 100 [2 ml, 100 ng cutoff (C.O.)]

Methamphetamine ng/100 cm^2	% C.O.	Positives(%)	Negatives (%)	Equivocal (%)
0	0	0	100	0
50	−50	90	10	0
75	−25	90	10	0
100	C.O.	100	0	0
125	+25	100	0	0
150	+50	100	0	0

LFIA 500 [10 ml, 500 ng cutoff (C.O.)]

Methamphetamine ng/100 cm^2	% C.O.	Positives(%)	Negatives (%)	Equivocal (%)
0	0	0	100	0
250	−50	90	10	0
380	−25	90	10	0
500	C.O.	100	0	0
630	+25	100	0	0
750	+50	100	0	0

Process-Based Assessment of Decontamination of a Former Methamphetamine Laboratory in a Hotel Room

Methamphetamine surface contamination was confirmed by the colorimetric method, LFIA (50 and 500 ng/100 cm^2) methods and LC/MS (NMAM 9111) during the initial visit. A map of the room indicating locations and levels of contamination was prepared but, not provided to the contractor hired by hotel management to clean the room. Recommendations were made as to what materials needed to be discarded (carpet, lamp shades, room air conditioner filters,

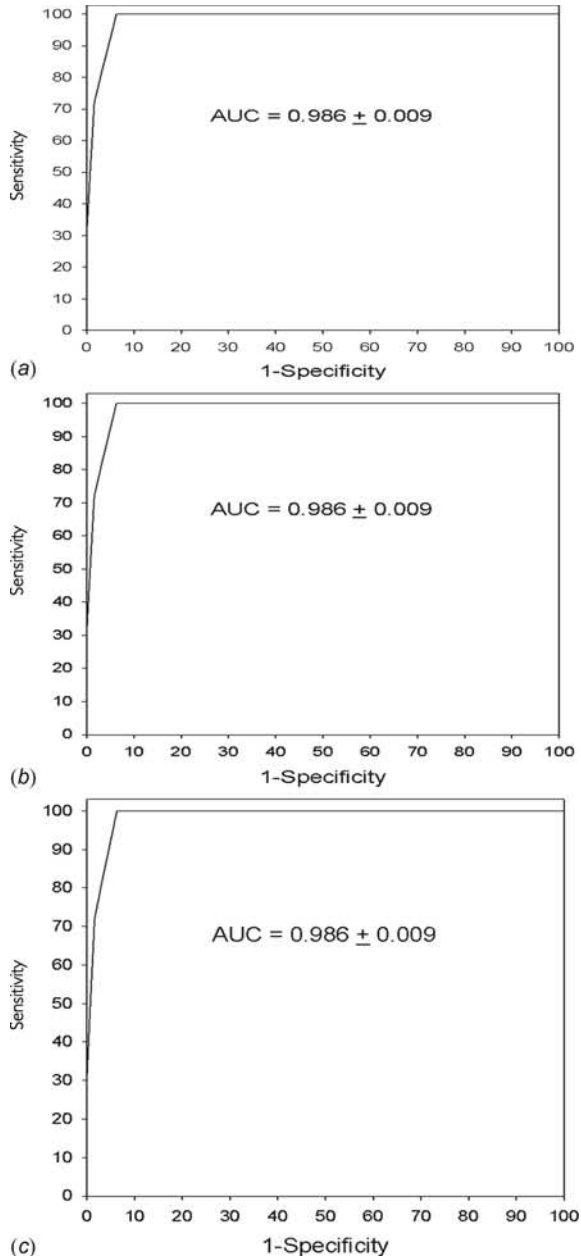

FIG. 3—*ROC curves based on LFIA surface wipe tests. Sensitivity, or the true-positive fraction, was plotted on the Y axis. The false-positive fraction (or 1- specificity) was plotted on the X axis. Accuracy was measured by calculating the area under the ROC curve (AUC). An area of 1 represents an ideal test; values near 0.5 represent an indiscriminant test. A = 50 ng LFIA, B = 100 ng LFIA and C = 500 ng LFIA.*

TABLE 3—*Comparison of LFIA (50, 100, and 500) surface detection method to Draft Method 9111, LC–MS with Isotopic Dilution.*

Methamphetamine	LFIA 50	Draft 9111
0	Negative (9/9)	ND
38 ng/100 cm^2	Positive 9/9	Below reporting limit (<100 ng/100 cm^2)
50 ng/100 cm^2	Positive 9/9	Below reporting limit (<100 ng/100 cm^2)
63 ng/100 cm^2	Positive 9/9	Below reporting limit (<100 ng/100 cm^2)
Methamphetamine	LFIA 100	Draft 9111
0	Negative (9/9)	ND
75 ng/100 cm^2	Positive 9/9	Below reporting limit (<100 ng/100 cm^2)
100 ng/100cm^2	Positive 9/9	94 ± 7 ng/100 cm^2
125 ng/100 cm^2	Positive 9/9	118 ± 4 ng/100 cm^2
Methamphetamine	LFIA 500	Draft 9111
0	Negative (9/9)	ND
380 ng/100 cm^2	Positive 9/9	400 ± 11 ng/100 cm^2
500 ng/100 cm^2	Positive 9/9	490 ± 12 ng/100 cm^2
630 ng/100 cm^2	Positive 9/9	581 ± 31 ng/100 cm^2

and grills), laundered (drapes and bed linen) or cleaned in place (tables, desks, etc.,). Samples from the same locations as the initial visit were taken during the remediation and a second contamination map prepared. The cleaning technicians were advised to change their cleaning techniques, remove or replace certain fixtures and provided with training and a supply of the LFIA tests to perform themselves. The final inspection after further remediation efforts found all but one of the contaminated locations were below the limits of detection (Table 4) of the LFIA 50. Based on these results, Hamilton County (OH) Public Health released the room for occupancy.

Assessment of Methamphetamine Transfer From a Contaminated Former Methamphetamine Laboratory to Personal Property Brought into the Residence

Initial assessment of the residence with LFIA qualitative tests revealed extensive but, relatively low levels of surface contamination throughout the residence (positive LFIA 50 and LFIA 500). Horizontal architectural surfaces (window sills, tops of moldings, cabinets, shelves, etc.,) were found to have higher levels than vertical surfaces. Based on demonstrated surface contamination and interviews with law enforcement and the owner of the property, the location was declared a confirmed former methamphetamine laboratory and was determined by Hamilton County (OH) Public Health to be unsafe. Residents were relocated, leaving behind nearly all of their personal property. Quantitative surface wipe samples were collected and analyzed by Draft NMAM 9111. The presumed location of methamphetamine manufacturing was determined to have occurred in the basement (highest contamination = 18 μg/100 cm^2) and methamphetamine

TABLE 4—*Assessment of performance of LFIA 50, and LFIA 500 in comparison to LCMS (Draft 9111) to determine decontamination of a clandestine laboratory located in motel room.*

	Pre-Remediation			During-Remediation			After Remediation		
Location	LFIA 50	LFIA 500	LCMS (μg/100 cm^2)	LFIA 50	LFIA 500	LCMS (μg/100 cm^2)	LFIA 50	LFIA 500	LCMS (μg/100 cm^2)
Dresser A	POS	POS	11.00	POS	NEG	0.34	NEG	NEG	ND*
Dresser B	POS	POS	14.00	POS	NEG	0.07	NEG	NEG	ND
TV Stand	POS	POS	12.00	POS	POS	0.40	NEG	NEG	ND
TV	POS	POS	4.80	NEG	NEG	ND*	NEG	NEG	ND
AC vent	POS	POS	24.00	POS	POS	1.20	NEG	NEG	ND
AC return	POS	POS	26.00	POS	POS	3.20	NEG	NEG	ND
Wall	POS	POS	4.20	POS	POS	0.89	POS	NEG	ND
Table	POS	POS	1500.00	POS	POS	4.80	NEG	NEG	ND
Window	POS	POS	2.10	NEG	NEG	ND*	NEG	NEG	ND
Night stand	POS	POS	5.50	POS	NEG	0.07	NEG	NEG	ND
Drapes	POS	NEG	0.78	POS	NEG	0.13	NEG	NEG	ND

* ND = Not Detected

was found on surfaces throughout the first (highest contamination = 6.7 μg/100 cm^2) and second floors (highest contamination = 3.2 μg/100 cm^2). The furnace and HVAC ducts were contaminated and could have been an initial source of distributing methamphetamine.

To assess the levels of transfer of methamphetamine from contamination inherent to the building onto personal property, representative items were tested with 50 and 500 ng LFIA. Items that had direct physical contact with contaminated building surfaces, such as an item placed on a shelf, were frequently contaminated. Other items were found to have contamination from probable transfer by persons handling an object after coming in contact with a contaminated surface. For example, opening a window with contamination on the sill, may lead to the person transferring a portion of that contamination to the next object they handled. In all, nearly 240 tests were used to test personal property in the home. Items that were below the detection limit of the 50 ng LFIA were released immediately to the former tenants. Items that tested positive at 50 ng but, were below the detection limit of the 500 ng LFIA were cleaned by wiping with a sanitizing wipe containing quaternary ammonia, retested at 50 ng and if below the limit of detection, returned to the former tenant. Items that tested positive at the 500 ng LFIA and would cost more than one hundred dollars to replace were decontaminated if possible. Usually, a single application of a foaming cleaner containing quaternary ammonia followed by removal with a blotting motion using clean paper towels resulted in subsequent negative tests using the 50 ng LFIA. Hamilton County (OH) Public Health ultimately worked with the property owner to develop a process-based decontamination plan to return the

property to a habitable status. Through a process of cleaning, testing, cleaning and re-testing, the property eventually was declared cleared for habitation.

Thousands of illicit drug laboratories are found each year. While active methamphetamine laboratories represent a significant source of toxic or potentially lethal chemical exposures [8] a majority of clandestine labs are identified long after they cease activity. Residual contamination of these clandestine laboratories represents a hazard to thousands of emergency response personnel, remediation workers and the general public. Researchers from the NIOSH developed rapid, sensitive surface sampling technologies to assess the location and level of methamphetamine contamination in clandestine labs. Two methods were developed, a colorimetric test and an immunochemical test. These technologies were developed in tandem to do initial confirmation of the presence of methamphetamine on surfaces using the colorimetric method and then use the immunochemical method as a sensitive semi-quantitative detection method to determine the extent of contamination and assess remediation effectiveness. This method is also suitable to detect contamination on personnel and equipment. The tests are simple, rapid, accurate and relatively inexpensive.

References

[1] Nonnemaker, J., Engelen, M., and Shive, D., "Are Methamphetamine Precursor Control Laws Effective Tools to Fight the Methamphetamine Epidemic?," *Health Econ.*, Vol. 20, No. 5, 2010, pp. 519–531.

[2] McBride, D. C., Terry-McElrath, Y. M., Chriqui, J. F., O'Connor, J. C., and VanderWaal, C. J., "The Relationship Between State Methamphetamine Precursor Laws and Trends in Small Toxic Lab Seizures," 2008, *Document No. 223467*, National Institute of Justice, US Department of Justice, Washington, D.C., 2008.

[3] O'Connor, J. C., Chriqui, J. F., and McBride, D. C., "Developing Lasting Legal Solutions to the Dual Epidemics of Methamphetamine Production and Use," *N D Law Rev.*, Vol. 82, No. 165, 2006, pp. 1165–1194.

[4] National Drug Threat Assessment, *Document No. 2008-Q0317–005*, National Drug Intelligence Center, US Department of Justice, Washington, D.C., 2009.

[5] Burgess, J. L., Barnhart, S., and Checkoway, H., "Investigation of Clandestine Drug labs: Adverse Medical Effects in Law Enforcement Personnel," *Am. J. Ind. Med.*, Vol. 30, 1996, pp. 488–494.

[6] Cooper, D., Souther, L., Hanlon, D., et al., "Public Health Consequences Among First Responders to Emergency Events Associated With Illicit Methamphetamine Laboratories—selected states, 1996–1999," *MMWR Morb Mortal Wkly Rep.*, Vol. 49, 2000, pp. 1021–1024.

[7] Thrasher, D. L., Von Derau, K., and Burgess, J., "Health Effects From Reported Exposure to Methamphetamine Labs: A Poison Center-Based Study," *J. Med. Toxicol.*, Vol. 5, No. 4, 2009, pp. 200–204.

[8] Martyny, J., Arbuckle, S., McCammon, C. Jr., Esswein, E., Erb, N., and Van Dyke, M., "Chemical Concentrations and Contamination Associated with Clandestine Methamphetamine Laboratories," *J.Chem. Health Safety*, Vol. 14, No. 4, 2007, pp. 40–52.

[9] VanDyke, M., Erb, N., Arbuckle, S., and Martyny, J., "A 24-hour Study to Investigate Persistent Chemical Exposures Associated With Clandestine Methamphetamine Laboratories," J. Occup. Environ. Hyg., Vol. 6, No. 2, 2009, pp. 82–89.

[10] Martyny, J., Arbuckle, S., McCammon, C. Jr., Erb, N., and Van Dyke, M., "Methamphetamine Contamination on Environmental Surfaces Caused by Simulated Smoking of Methamphetamine," *J. Chemical Health and Safety*, Vol. 15, No. 5, 2008, pp. 25–31.
[11] Grant, P. J. "Evaluation of Children Removed From a Clandestine Methamphetamine Laboratory," *J. Emerg. Nurs.*, Vol. 33, 2007, pp. 31–41.
[12] Smith, J. P., Martin, A., Sammons, D. L., Striley, C., Biagini, R., Quinn, J., Cope, R., and Snawder, J. E., "Measurement of Methamphetamine on Surfaces Using Surface Plasmon Resonance," *Toxicol. Mech. Methods*. Vol. 19, No. 6–7, 2009, pp. 416–421.
[13] NIJ Standard–0604.01, "Color Test Reagents/Kits for Preliminary Identification of Drugs of Abuse," National Institute of Justice, Office of Science and Technology, Washington, D.C., 2000.
[14] Nakaharab, Y., and Sekinea, H., "A High Selective Screening Test for Methamphetamine in Human Urine," *Forensic Sci. Int*., Vol. 26, No. 4, 1984, pp. 277–282.

Author Index

Subject Index

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ISBN: 978-0-8031-7519-8
Stock #: STP1533