Environmental Sampling in Healthcare Settings
Dr. Lynne Sehulster, CDC
A Webber Training Teleclass

Environmental Sampling
in Healthcare Settings:
Introduction to Basic Principles

Lynne Sehulster, PhD, M(ASCP)
Division of Healthcare Quality Promotion
Centers for Disease Control and Prevention

Hosted by Paul Webber
paul@webbertraining.com
www.webbertraining.com

Objectives for Today’s Presentation

• Introduction to environmental sampling principles and practices for indoor healthcare settings
• Air
  – General methods
  – Microbiology vs. particle counts
• Surfaces
  – General methods
  – Environmental surfaces, instruments and devices, hands
• Water
  – General methods
  – Ensuring accurate and meaningful results

CDC’s “EIC” Guideline 2003

• Environmental Services
• Environmental Sampling

Where Can I Find the EIC Guidelines?

• Part II Recommendations:
  • MMWR 2003; 52 (RR-10): 1-44
  • Errata: MMWR 2003; 52 (42): 1025-6
• Full text version:
  • www.cdc.gov/nicdd/dhsp/gp/gp_environinfection.html
• Print version (ASHE):
  • www.hospitalconnect.com/ashe/resources/Importantresources.html

Environmental Sampling Section

Hosted by Paul Webber paul@webbertraining.com
www.webbertraining.com
Environmental Sampling

Environmental microbiology is not clinical microbiology
Sampling is supported by epidemiologic assessment
Random, undirected sampling is not recommended
Sampling requires a protocol for sampling and culturing, analysis of results, and action based on the interpretation of results

Environmental Sampling

Suggested uses:
- Support for outbreak investigation when epi suggests environmental reservoirs or fomites are implicated in disease transmission
- Research in environmental infection control
- Monitor a potentially hazardous situation
- Evaluate a change in environmental infection control for quality assurance purposes
- Perform periodic regular maintenance of equipment (i.e., HVAC systems)
- Legal issues

Current routine monitoring:
- Biological monitoring of sterilization processes
- Monthly culturing of water used in hemodialysis applications and for final dialysate use dilution

Environmental Sampling

Expensive and time-consuming; subject to many variables in protocol, analysis, and interpretation

“Sampling is a public exercise and is always subject to disclosure; therefore, the investigator is required to minimize false negatives and, more rarely, false positives.”


Air Sampling

Primarily to determine bacteria and fungi identities and concentration in biological aerosols

Major methods:
- Impingement in liquids
- Impaction on solid surfaces
- Sedimentation (e.g., settle plates)

Requires an understanding of what is being measured and a full description of the circumstances during sampling

Factors to Consider Prior to Conducting Microbiological Air Sampling

Conditions of the aerosol: particle size, amount of inert material, microorganism concentration, environmental factors
Sampling process: type of samplers and method, numbers of samples, duration of collection/sampling
Assay method to optimize microorganism recovery
Proper transport conditions to ensure sample quality and viability

Types of Air Samplers*

A. Impactor sampler (www.integraenv.com)
B. Glass impinger sampler (www.ubifcoa.com)
C. Slieve Impactor sampler (www.beamco.com)

* Examples for illustration purposes, not for endorsement

Hosted by Paul Webber  paul@webbertraining.com
www.webbertraining.com
Environmental Sampling In Healthcare Settings
Dr. Lynne Sehulster, CDC
A Webber Training Teleclass

Compare and Contrast the Main Air Sampling Methods

<table>
<thead>
<tr>
<th>Method</th>
<th>Principle</th>
<th>Suitable for Measuring</th>
<th>Collection Media or Surface</th>
<th>Points to Consider</th>
</tr>
</thead>
<tbody>
<tr>
<td>Impingement in liquid</td>
<td>Air drawn in through small jet, directed against liquid surface</td>
<td>Visible microorganisms, water aerosols</td>
<td>Liquid particle, particle size, concentration, time</td>
<td>Used for liquid particle sampling</td>
</tr>
<tr>
<td>Impingement on solid surface</td>
<td>Air drawn into sampler, particles deposited on dry surface</td>
<td>Visible particles, visible microorganisms</td>
<td>Dry surfaces, coated surfaces, film</td>
<td>Used for bacteria and fungal spore sampling, high volume can be sampled</td>
</tr>
<tr>
<td>Sedimentation (settling)</td>
<td>Particles and microorganisms settle as gravity</td>
<td>Visible particles, visible microorganisms</td>
<td>Nutrient agar in plates or slides</td>
<td>Simple, best suited for qualitative sampling, not used for fungal spores</td>
</tr>
</tbody>
</table>

Before You Do Microbiological Air Sampling...
- Define your objective and analytical approach
  - Qualitative vs. quantitative
- Concentration over time? Particle size?
- Compare to counts from outdoor air
- Understand that results will not reflect “real time”
- Fully describe the circumstances in the area where sampling is occurring
- High volume sampling most efficient

Unresolved Issues and Microbiologic Air Sampling
- Unknown incubation period for IPA
- Infectious dose for Aspergillus spp. is unknown
- Lack of standard sampling protocols
- No standards or action levels for results

Particle Sampling
- Simple to perform, immediate results
- Verify HVAC system performance:
  - Filtration efficiency
  - Rank order from “dirty” to “clean”
- Verify infection control measures during construction:
  - Construction barrier and dust containment

Examples of Particle Counters

Source for both pictures: www.coastalsafety.net
Pictures are for illustration purposes only; not for endorsement

Hosted by Paul Webber  paul@webbertraining.com
www.webbertraining.com
Environmental Sampling In Healthcare Settings
Dr. Lynne Schulster, CDC
A Webber Training Teleclass

Example of Air Particle Analysis Using Condensate Particle Counter

<table>
<thead>
<tr>
<th>Location</th>
<th>Particles/c</th>
<th>Pressure (Pascal)</th>
<th>Filter Efficiency</th>
</tr>
</thead>
<tbody>
<tr>
<td>Outside</td>
<td>8500</td>
<td>na</td>
<td>- - - -</td>
</tr>
<tr>
<td>Lobby</td>
<td>1500</td>
<td>na</td>
<td>90 %</td>
</tr>
<tr>
<td>BMT Area Corridor</td>
<td>450</td>
<td>(+) 4 – 10</td>
<td>99.97 %</td>
</tr>
<tr>
<td>21 of 32 Rooms (BMT)</td>
<td>&lt; 10</td>
<td>(+) 6.6 – 30</td>
<td>99.97 %</td>
</tr>
<tr>
<td>9 of 32 Rooms (BMT)</td>
<td>80 – 80</td>
<td>(+) 6.6 – 17</td>
<td>99.97 %</td>
</tr>
<tr>
<td>2 of 32 Rooms (BMT)</td>
<td>&gt; 500</td>
<td>(+) 11 &amp; 12</td>
<td>99.97 %</td>
</tr>
<tr>
<td>Radiation Therapy</td>
<td>1500</td>
<td>na</td>
<td>90 %</td>
</tr>
<tr>
<td>Adjacent Building</td>
<td>5300</td>
<td>na</td>
<td>50 %</td>
</tr>
</tbody>
</table>

Source: A. Streiff & J. Widmer, AHAlka 2004

Reasons to Sample Air

- Preoccupancy verification of ventilation and cleanliness
- Establish baseline data (based on particle removal)
- BMTU, OR’s, NICU, other critical areas
- Post infection evaluation (outbreak)?
  - Verification of baseline data
  - Rule out ventilation as a source
  - Discover source of infectious fungi (reservoir?)
- Routine surveillance
  - CDC: not recommended without purpose
  - Some methods provide assurance of status quo
  - May be useful for finding deviations in baseline data

Source: A. Streiff & J. Widmer, AHAlka 2004

Baseline Data Development in Healthcare Air Sampling

- Provides verification of filtration efficiency
  - Should show relative drop of viable/non-culture particles
- Provides micro-flora verification in affected space
  - Air is not sterile; should reflect isolates similar to outside
- Baseline should compare data from indoor space and outdoors
- Baseline data is best established pre-occupancy
  - Ventilation systems should be working according to specs
  - Testing should be finished and specified ventilation parameters assured
- Ideally sampling should be conducted before occupancy to avoid variables.

Source: A. Streiff & J. Widmer, AHAlka 2004

Environmental Surface Sampling

- Decision to sample should be driven by epidemiology, infection control
- Disinfectant neutralizers may be needed
- Major methods include:
  - Sample/rinse using sponges, wipes or swabs
  - Direct immersion
  - Containment (interior surfaces of a container)
  - RODAC plate (direct surface sampling)

Things to Consider Prior to Surface Sampling

- Background – literature and present activities
  - Preliminary results from epidemiological investigation
- Locations to sample
- Collection method and equipment
- Number of replicate samples needed
- Are controls or comparisons needed?
- Parameters for assay; qualitative, quantitative, or both?
- Estimate of maximum allowable microbial numbers or types on surface(s) sampled
- Some anticipation of a plan of action based on results

Source: A. Streiff & J. Widmer, AHAlka 2004

Compare and Contrast Surface Sampling Methods

<table>
<thead>
<tr>
<th>Sample Type</th>
<th>Description</th>
<th>Target</th>
<th>Uses</th>
<th>Biological Agents</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wipe</td>
<td>Sterile 2 x 2 non-cotton gauze, incorporated, wipe area of known size</td>
<td>Nonporous surface, usually small or in area not easily accessible</td>
<td>Screening small nonporous surfaces, extent of contamination, decontamination effectiveness</td>
<td>Bacteria, viruses, fungi, biological toxins</td>
</tr>
<tr>
<td>Swab</td>
<td>Sterile non-cotton swabs, individually packaged, then moistened with sterile solution; wipe area of known size</td>
<td>Nonporous surface, usually small or in area not easily accessible</td>
<td>Screening small nonporous surfaces, extent of contamination, decontamination effectiveness</td>
<td>Bacteria, viruses, fungi, biological toxins</td>
</tr>
<tr>
<td>RODAC</td>
<td>Convergent agar surface in culture dish, pressed onto surface, inoculated</td>
<td>Nonporous surfaces, relatively small area</td>
<td>Screening small nonporous surfaces, extent of contamination, decontamination effectiveness</td>
<td>Bacteria, fungi</td>
</tr>
</tbody>
</table>


Hosted by Paul Webber  paul@webbertraining.com
www.webbertraining.com
Environmental Sampling In Healthcare Settings
Dr. Lynne Sehulster, CDC
A Webber Training Teleclass

**Things to Consider Before Conducting Surface Sampling**
- Asepsis is critical
  - Sterilized sampling materials
  - Aseptic technique
- Document the circumstances of sampling
  - State of the surface and its preparation, if any, prior to sampling
- Prepare a sampling strategy or plan that ensures the validity of the results and is appropriate for the organism(s) being sampled

**Wipe Method**
- Materials used:
  - Sterile gloves, sterile sample containers, sterile wrapped 2x2 gauze sponge pads, disposable sterile sampling template, sterile water or other appropriate fluid, plastic bags, identification tags
- Affix the template
- Aseptically wet the gauze with fluid and thoroughly wipe the area within the template
- Fold the gauze so the exposed side is inward and place in sample container; label
- Repeat with new template and new gauze if another surface is to be sampled

**Swab Sampling Procedure**
- Materials used:
  - Sterile items: gloves, sample containers (e.g., large vials), wrapped non-cotton swabs, wetting solution, scissors, disposable template
  - Sealable plastic bags, identifying markers, tags
- Affix the template to the surface
- Wet the swab and wipe using an S-shaped pattern (vertically & horizontally), rolling the swab over the surface
- Place the swab aseptically in a sample tube; label
- Change gloves and use a new template if sampling another surface

**RODAC Plate Sampling Method**
- Materials used:
  - RODAC plate (agar medium is overfilled to give a convex surface)
- Used to sample cleaned surfaces; not suitable for visibly dirty or irregular surfaces
- Neutralizers can be incorporated into the medium if surface disinfectant residuals are present
- Press the convex medium onto the surface; do not twist or move the plate around

**Neutralizing Agents**

<table>
<thead>
<tr>
<th>Disinfectant</th>
<th>Neutralizer or Neutralizing media</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sodium hypochlorite, chlorine</td>
<td>Sodium thiosulfite, Dey Engley</td>
</tr>
<tr>
<td>double, iodine</td>
<td>(D/E) broth or agar</td>
</tr>
<tr>
<td>Formamidine, gluteraldehyde</td>
<td>Glucose, D/E broth or agar</td>
</tr>
<tr>
<td>Hydrogen peroxide</td>
<td>Catalase</td>
</tr>
<tr>
<td>Phenolics</td>
<td>Tween 80, D/E broth or agar</td>
</tr>
<tr>
<td>Quaternary ammonium compounds</td>
<td>Lacticin + Lubrol WX, Lethane broth or agar, D/E broth or agar</td>
</tr>
<tr>
<td>Vaporized hydrogen peroxide</td>
<td>None needed – end products are H₂O and O₂</td>
</tr>
</tbody>
</table>


**Visual Methods Currently Used to Evaluate Cleaning**
- Application of clear chemicals that fluoresce under UV light
  - Glo Germ
  - Gluten-derived “glues” + detergent + fluorescent dye
  - Qualitative: Yes / No
- ATP
  - Proprietary swabs and solution
  - Luminometer reads presence of organic matter - expressed in relative light units (RLU)
  - Can be quantitative

Hosted by Paul Webber paul@webbertraining.com
www.webbertraining.com
Environmental Sampling In Healthcare Settings
Dr. Lynne Sehulster, CDC
A Webber Training Teleclass

Cleaning in the ICU: The Most and the Least

Water Sampling
- Sampling “finished” water often requires the use of a chlorine neutralizer (i.e., sodium thiosulfate)
- Use media and incubation temperatures appropriate for culture of “stressed” organisms (R2A, diluted peptone; ambient temperature)
- Specific methods are used for Legionella and other waterborne microorganisms

Considerations When Sampling for Legionella Spp.
- Point-of-use devices and system surfaces
- Faucet aerators and showerheads:
  - Swab surfaces of these fixtures first
  - Water samples are collected after the aerators and shower heads are removed
- Collect 1 L water samples in sterile containers
- Collected swabs should be immersed in 5 – 10 mL of water from the same device
- Suitable media for Legionella culture: BCYE (buffered charcoal yeast extract)

For More Details...
- CDC’s EIC Guideline

Thank You!
Division of Healthcare Quality Promotion
Centers for Disease Control and Prevention

“Protect patients, protect health-care personnel, and promote safety, quality, and value in the health-care delivery system”

Hosted by Paul Webber  paul@webbertraining.com
www.webbertraining.com
Environmental Sampling In Healthcare Settings
Dr. Lynne Sehulster, CDC
A Webber Training Teleclass

THE NEXT FEW TELECLASSES

<table>
<thead>
<tr>
<th>Date</th>
<th>Title</th>
<th>Speaker</th>
<th>Details</th>
</tr>
</thead>
<tbody>
<tr>
<td>25 Jun</td>
<td>South Dakota Teleclass: Pathoral Lab Update</td>
<td>Dr. Steve McDole, Aurora District Health Board</td>
<td></td>
</tr>
<tr>
<td>26 Jun</td>
<td>CDC Teleclass 2: The CDC Examination Process, Gomander update</td>
<td></td>
<td></td>
</tr>
<tr>
<td>17 Jul</td>
<td>Free Teleclass Community Associated MERSA: What's Up, What's Next</td>
<td>Dr. Rachel Zoberli, CDC</td>
<td></td>
</tr>
<tr>
<td>22 Jul</td>
<td>Free Teleclass Progress Report from the Chief Nursing Office</td>
<td>Christine Beasley, British Department of Health</td>
<td></td>
</tr>
<tr>
<td>7 Aug</td>
<td>Free Teleclass Disinfection &amp; Sterilization: Current Issues &amp; New Research</td>
<td>Dr. William Rutala, University of North Carolina</td>
<td></td>
</tr>
<tr>
<td>14 Aug</td>
<td>Free Teleclass Extended Spectrum Beta Lactamase and Infection Control</td>
<td>Prof. David Paterson</td>
<td></td>
</tr>
</tbody>
</table>

www.webbertraining.com/schedulep1.php

Hosted by Paul Webber paul@webbertraining.com
www.webbertraining.com