Microbiologic Sampling of the Environment

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Microbiologic Sampling of the Environment

Lecture Goals

Microbiologic Sampling

- Indications
- Methods
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History

- Pre-1970, hospitals regularly cultured air and surfaces
- By 1970, AHA advocated discontinuation because HAI not associated with levels of microbes in the air and surfaces; not cost-effective
- In 1981, CDC recommended targeted sampling (eg, sterilizers and dialysis water)
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CDC Guidelines for EIC, 2003

- Targeted microbiological sampling
  - Support of an investigation of an outbreak
  - Research
  - Monitor a potentially hazardous environmental condition
  - Quality assurance
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- Will environmental sampling provide meaningful, interpretable data that help identify actual or potential contamination problems associated with a specific procedure or instrument?
- Should not be done if no plan for interpreting and acting on the results obtained.
- Is it justified on epidemiological grounds?
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Investigation of an Outbreak

When?

- Environmental reservoirs or fomites are implicated epidemiologically in disease transmission
- Plan for interpreting and acting on the results
- Plan to link microorganisms from the environment with clinical isolates by molecular epidemiology
Outbreak: two patients in CT-ICU with symptomatic *B. cepacia*

Epidemiologic investigation: case-control study revealed that both patients required an intra-aortic balloon pump (IABP) for circulatory support

Microbiological investigation: water reservoir of IABP contained $>10^5$ *B. cepacia*/ml
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Investigation of an Outbreak

- Microbiologic investigation: causative organism isolated from several components of the IABP and the hands of a nurse who manipulated the IABP’s buttons/switches.
- Molecular epidemiology: similar plasmid profile from strains from the patients and the IABP.
- Conclusion: transmission presumably occurred during manipulation of IV lines.
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Research

- When? Experimental methods that provide new information about the spread of HAIs

- Example: Relation of the Inanimate Hospital Environment to Endemic Nosocomial Infection (NEJM 1982;302:1562).

- Cultured air, surfaces, and fomites in old/new hospital and despite major differences in contamination (17% positive vs 5%), incidence of NI remained unchanged.
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Monitor a Potentially Hazardous Environmental Condition

• When? Confirm the presence of a hazardous chemical/biological agent, and validate abatement of the hazard

■ Examples
  ◆ Detect bioaerosols (eg, ultrasonic cleaner)
  ◆ Detect agent of bioterrorism
  ◆ Sample for IH (eg, sick building)
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Quality Assurance

- When? To evaluate the effects of a change in infection control practice or ensure equipment/systems perform as expected
  - Air sampling during construction/renovation to qualitatively detect breaks in IC measures
  - Only routine sampling recommended: biological monitoring of sterilizers, monthly cultures of water used in hemodialysis
  - Endoscopes/AERs
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Air Sampling

General comments

- Particles in a biological aerosol usually vary from <1 to > 50 µm.
- Particles consist of a single, unattached organism or clumps.
- Vegetative cells do not ordinarily survive long in air.
- Pathogens may settle on surfaces and become airborne again with sweeping, etc.
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Air Sampling

- Air sampling for QA is problematic due to the lack of uniform air quality standards
- The critical number of *Aspergillus* that poses a risk for neutropenic patients is not known
- Results affected by factors (traffic, time of year)
- Results need to be compared to other defined areas
Air Sampler
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Air Sampling

- Impingement in liquids
- Impaction on solid surfaces
- Sedimentation
- Filtration
- Centrifugation
- Electrostatic precipitation
- Thermal precipitation
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Air Sampling

- Factors in Selecting an Air Sampling Device
  - Viability and type of organism
  - Skill required to operate sampler
  - Availability and cost of sampler
  - Availability of auxiliary equipment (vacuum pump)
  - Assumed concentration and particle size
  - Sensitivity of microorganisms to sampling
  - Compatibility with the selected method of analysis
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Air Sampling

- Impaction on solid surfaces (sieve)-collects (mo deposited on agar), sizes, conc per unit volume of air (CFU/ft$^3$). Ex. Aspergillus
- Impingement in liquids-collects (mo directed against a liquid [nutrient broth], conc over time) Ex. Water aerosols for Legionella
- Sedimentation (settle plates)-mo settle on agar via gravity, conc over time (CFU/time)
Filtering

Bacteria are captured by the filter.

Water passes through the filter.
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Water Sampling

- When? Routine testing of water not indicated (except dialysis) but sampling in support of outbreak investigation can help determine IC measures

- Use established methods (eg, sample water ASAP after collection, 100ml minimum, sterile collection equipment, neutralizers, recovery media and incubation temp [diluted peptone, 20°C], pour plates [high counts], membrane filtration-0.2µ [low counts, larger volumes])

- Filters are placed on agar plates and incubated for 48h
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Surface Sampling Methods

- Sample/Rinse-use sterile wipe/sponge/wipe, media, qualitative/quantitative assays
- Direct Immersion-immerse in media, then assay
- Containment-interior surfaces of containers
- RODAC (replicate organism detection and counting)-sampling flat, nonabsorbent surfaces, direct assay
Methods for Culturing Surfaces
CDC, 2003; Boyce, 2012

- Moistened swab
- Moistened swab and rinse (broth enrichment)
- Moistened sponge and rinse
- Moistened wipe and rinse
- Direct Immersion
- RODAC plates
- Irregular objects
- Irregular objects
- Irregular objects
- Large, flat surfaces
- Large, flat surfaces
- Immerse in broth
- Flat surfaces
Moistened Swab with Direct Plating

Boyce, 2012

- Use moistened swab to sample surfaces
  - If defined area not sampled, results are semi-quantitative
  - If defined area sampled using a template, results are quantitative (CFUs/cm²); preferable
- Moistening (wetting) agents include normal saline, broth media (neutralizers)
- Swab is used to directly inoculate non-selective or selective media, followed by incubation x 48h
- Use for sampling irregular-shaped objects
Moistened Swab with Direct Plating
Boyce, 2012

- Advantages
  - Easy to perform
  - Simple; can be used in many facilities with microbiology laboratory support
  - Provides information about general level of contamination or for specific pathogens

- Disadvantages
  - Least sensitive method for detecting or organisms on surfaces
  - Non-standardized procedure makes comparison of studies difficult
RODAC Plates
Boyce, 2012

- Small petri plate filled with agar to provide convex surfaces
- Agar plate is pressed against a flat surface, plate is incubated
- Advantages: very easy to perform and standardized; results expressed as CFU/cm²; neutralizer available
- Disadvantages: greater cost; sample small area per plate
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Surface Sampling

- Used for research (potential reservoirs of pathogens, survival of microorganisms on surfaces, source of contamination), as part of an epidemiologic investigation, or QA purposes
- Media (nutrient-rich such as TSA or BHI), reagents, and equipment required for surface sampling available in micro lab
- Effective sampling requires moisture
Other Microbiologic Sampling

- Biological indicators
- Hemodialysis water - 200/ml, 2000/ml
- Infant formula-hospital prepared
- Pharmacy-hospital prepared
- Respiratory therapy
- Blood bank water bank-used to thaw plasma
- Endoscopes
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Conclusions

- Do not conduct random microbiological sampling of air, water, and surfaces
- When indicated, conduct microbiologic sampling as part of an epidemiologic investigation
- Limit microbiologic sampling for QA to: biological monitoring, dialysis water, or evaluation of IC measures
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Reference