Laboratory Workers Beware: Laboratory Acquired Infections

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Case report

- 35 yo male
- Presented to ED with acute malaise, fever and myalgias
- Discharged on oral antibiotics
- Returned next day with worsening symptoms
- Developed tachycardia and hypotension
- Died 3 hours later
- Worked in a hospital microbiology laboratory
Case report

- Blood cultures subsequently grew *N. meningitidis* serogroup C
- He had processed positive blood culture from a patient with *N. meningitidis* serogroup C 3 days prior to symptoms onset
- Isolates indistinguishable by PFGE and MLEE
- Positive blood culture bottles were not processed in a safety cabinet

MMWR 51:141
## Epidemiology of Laboratory Acquired Infections

<table>
<thead>
<tr>
<th>Year</th>
<th>Location</th>
<th>Person-years</th>
<th>No. of infections</th>
<th>Annual incidence per 100,000 Lab Workers</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>1984</td>
<td>Utah, USA</td>
<td>5955</td>
<td>18</td>
<td>302</td>
<td>JCM 21:486</td>
</tr>
<tr>
<td>1986</td>
<td>Minnesota, USA</td>
<td>2290</td>
<td>8</td>
<td>350</td>
<td>Am J Pub Health 78:1213</td>
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<tr>
<td>1986</td>
<td>USA</td>
<td>4202</td>
<td>6</td>
<td>140</td>
<td>Am J Pub Health 78:1213</td>
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<tr>
<td>1986</td>
<td>UK</td>
<td>28524</td>
<td>15</td>
<td>52.6</td>
<td>J Clin Path 42:677</td>
</tr>
<tr>
<td>1989</td>
<td>UK</td>
<td>21756</td>
<td>18</td>
<td>82.7</td>
<td>J Clin Path 44:667</td>
</tr>
<tr>
<td>1995</td>
<td>UK</td>
<td>55698</td>
<td>9</td>
<td>16.2</td>
<td>J Clin Path 52:415</td>
</tr>
</tbody>
</table>
Epidemiology of Laboratory Acquired Infections

<table>
<thead>
<tr>
<th>Table 1—Summary of Laboratory-Acquired Infections, 1930-1963</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Type of Infection</strong></td>
</tr>
<tr>
<td></td>
</tr>
<tr>
<td>----------------------</td>
</tr>
<tr>
<td>Bacterial</td>
</tr>
<tr>
<td>Viral</td>
</tr>
<tr>
<td>Rickettsial</td>
</tr>
<tr>
<td>Fungal</td>
</tr>
<tr>
<td>Parasitic</td>
</tr>
<tr>
<td><strong>Totals</strong></td>
</tr>
</tbody>
</table>

* Sulkin and Pyle.†
‡ Plague.
‡ Nine of these were B virus; two were Russian spring-summer encephalitis.
§ Toxoplasmosis.
Epidemiology of Laboratory Acquired Infections

Table 1. Ten most frequently reported laboratory-associated infections worldwide.

<table>
<thead>
<tr>
<th>Disease</th>
<th>No. of cases</th>
<th>No. of deaths</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brucellosis</td>
<td>426</td>
<td>5</td>
</tr>
<tr>
<td>Q fever</td>
<td>280</td>
<td>1</td>
</tr>
<tr>
<td>Hepatitis</td>
<td>268</td>
<td>3</td>
</tr>
<tr>
<td>Typhoid fever</td>
<td>258</td>
<td>20</td>
</tr>
<tr>
<td>Tularemia</td>
<td>225</td>
<td>2</td>
</tr>
<tr>
<td>Tuberculosis</td>
<td>194</td>
<td>4</td>
</tr>
<tr>
<td>Dermatomycoses</td>
<td>162</td>
<td>0</td>
</tr>
<tr>
<td>Venezuelan equine encephalitis</td>
<td>146</td>
<td>1</td>
</tr>
<tr>
<td>Psittacosis</td>
<td>116</td>
<td>10</td>
</tr>
<tr>
<td>Coccidioidomycosis</td>
<td>93</td>
<td>2</td>
</tr>
</tbody>
</table>

**NOTE.** Data are for the years 1976 [3] and 1978 [4].
Epidemiology of Laboratory Acquired Infections

Risk of a laboratory-acquired infection in microbiologists versus the general population of the same relative age

<table>
<thead>
<tr>
<th>Organism</th>
<th>Risk/100,000 microbiologists</th>
<th>Risk/100,000 general population</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brucella</td>
<td>641</td>
<td>0.08</td>
</tr>
<tr>
<td>Coccidioides</td>
<td>13.7</td>
<td>12</td>
</tr>
<tr>
<td>C. difficile</td>
<td>0.2</td>
<td>8</td>
</tr>
<tr>
<td>E. coli O157:H7</td>
<td>8.3</td>
<td>0.96</td>
</tr>
<tr>
<td>N. meningitidis</td>
<td>25.3</td>
<td>0.62</td>
</tr>
<tr>
<td>Salmonella</td>
<td>1.5</td>
<td>17.9</td>
</tr>
<tr>
<td>Shigella</td>
<td>6.6</td>
<td>6.6</td>
</tr>
</tbody>
</table>
Brucella sp.

- Cattle (B. abortus)
- Swine (B. suis)
- Goats and sheep (B. melitensis)
- Dogs (B. canis)
- Milk
- Slaughter
- Veterinary care
- Vaccination accident or laboratory accident, with cultures
Brucella sp.

- 20-50 cases reported in Australia annually (majority in Qld)
- Most commonly reported laboratory acquired infection
- 24% of cases, 11% of deaths
- Acquisition
  - Aerosolisation of cultures
  - Direct contact with cultures
  - Often cannot be determined
Brucella sp.

- Reported incidents:
  - Breakage of centrifuge tubes, BC bottles
  - Sniffing plates
  - Manipulating colonies on open bench
  - Not using PPE
  - Mouth pipetting
  - Unknown

- Attack rate: 30-100%

- Misidentification a major factor
Brucella sp

- Report from Canada:
  - *Brucella melitensis* isolate from draining abscess initially misidentified as *Moraxella phenylpyruvica* by API
  - All work carried out on open bench
  - Identified as Brucella 22 days after culture collected
  - 6 staff directly manipulated organism on open bench
  - All offered prophylaxis; one declined
    - Doxy 100mg bd, Rif 600mg od for 3/52
  - Clinical brucellosis developed in the lab worker who did not receive prophylaxis 10 weeks later
Brucella sp

- Post exposure recommendations
  - Risk stratify
    - High:
      - Manipulated colonies on an open bench (or within 5’)
      - in room when aerosol generating procedure conducted
      - sniffed plates
      - direct skin contact,
      - spray into eyes/nose/mouth
      - Inoculation
      - mouth pipetting
    - Low: All other staff in room not classified as high risk
Brucella sp

- Post exposure recommendations
  - Offer high-risk individuals prophylaxis
    - Doxy 100mg bd, Rif 600mg od for 3/52
    - Bactrim DS 1 bd for 3/52
  - Serological follow-up (high & low risk)
    - 0, 2, 4, 6 and 24 weeks (CDC)
    - 0, 6, 24 weeks for high risk (HPA)
    - Store baseline serum for low risk (HPA)
  - Monitoring for development of symptoms

J Clin Pathol 63:90–92
http://www.cdc.gov/nczved/divisions/dfbmd/diseases/brucellosis/recommendations.html
Brucella sp

Preventing laboratory exposure

- **Suspect Brucella**
  - Encourage requesting Drs to supply clinical information
  - Encourage laboratory staff to read clinical information (eg: ?Brucellosis, fever in a returned traveller, pig shooter)
  - Phenotypic characteristics
- Seal plates from patients with possible Brucella (eg: fever in returned traveller)
- Examine cultures from sterile sites inside safety cabinet
- Perform aerosol generating procedures on all organisms inside a safety cabinet
- Do not handle plates from sterile sites outside of a safety cabinet until organisms are identified with certainty
Neisseria meningitidis

- 200-300 cases in Australia each year
- Relative risk of invasive meningococcal disease in lab workers compared with general population ≈50
- Case fatality rate 50%
- All 16 reported cases occurred in laboratory staff working with sterile site isolates (not pharyngeal isolates)
- 15/16 reported cases worked with colonies on an open bench

J Clin Microbiol 2005; 43:4811
Neisseria meningitidis

Preventing Laboratory Exposure:
- Vaccine available for serogroups A, C, W135 and Y
  - 12% of reported cases in 2008 were from one of these serogroups (77% from serogroup B)
- Seal plates from patients with possible *N. meningitidis* (Gram negative diplococci seen)
- Examine cultures from sterile sites inside safety cabinet
- Perform aerosol generating procedures on all organisms inside a safety cabinet
- Do not handle plates from sterile sites outside of a safety cabinet until organisms are identified with certainty
- Use PPE
- Hand hygiene
- Consider prophylaxis if aerosol generating procedures inadvertently performed on *N. meningitidis* outside of a safety cabinet
Case

- 81 yo Female, from Philippines, in Australia since Oct 2010
- Left knee pain Dec 2009
- Worsening pain over 12 months. No systemic symptoms
- Investigated in Manila – aspirates - unhelpful
- Referred by GP to orthopaedics via ED
- Chronic discharge at previous aspiration sites
Case

Coronal

Bone erosion

Sagittal

phlegmon
Case

- Operative debridement in OT
  - Planned for subsequent TKR
- Histopathology:
Case

- Meanwhile in the micro lab….
  - Fungal culture plates growing a non-descript white mould at 48 hours
  - Subcultured outside of safety cabinet
  - One minute later – Histopathologist appears in the micro lab
    - DNA extraction performed in safety cabinet
      - Confirmed as *Coccidioides immitis* by ITS sequencing
      - Culture plates retrieved and destroyed
  - Further history: patient had lived in California for many years
Coccidioidomycosis

- Dimorphic fungus
- Acquired by inhalation of arthroconidia
- Half of cases subclinical
- Pneumonia, Cutaneous infection, bone/joint infection, meningitis

Ann NY Acad Sci 1111:315
Coccidioidomycosis

http://www.pfddb.net/photo/makimura_k/box0/standard/cocci_map.jpg
Coccidioidomycosis

- Infectious dose as low as one condidia
- Exposure in laboratory occurs when arthroconidia escape from a sealed culture plate if opened outside of a safety cabinet
- Degree of risk depends on age of culture (arthroconidia may develop from day 4)
- Laboratory exposures often more intense than natural acquisition, potentially worse disease
- Significant exposures: evacuation and decontamination of laboratory
- Prophylaxis for exposed laboratory staff recommended (and store baseline serum).
  - fluconazole or itraconazole 400mg od for 6 weeks and cease at 6 weeks if no seroconversion
  - Symptomatic monitoring for 6 months

Clin Inf Dis 49:919
Dimorphic fungi

- *Histoplasma, Blastomyces, Coccidioides, Paracoccidioides*

Preventing laboratory exposure

- **Suspect Dimorphic fungi**
  - Encourage requesting Drs to supply clinical information
  - Encourage laboratory staff to read clinical information (eg: Histoplasmosis)
  - Slow growing yeast from sterile site
  - Mould from a sterile site

- Seal fungal culture plates, examine in safety cabinet

- Do not handle plates of moulds outside of a safety cabinet until organisms are identified with certainty
Case

- 35 yo woman, previously well
- 6 day history of bloody diarrhoea, fever, headache and myalgias
- Commenced on IV ampicillin
- Blood and stool cultures grew *Salmonella* Typhi
- Subsequently developed hypotension, pulmonary odema, DIC
- Chloramphenicol and Gentamicin added
- Died 2 days after admission
Case

- 3 days after her death, her 14 year old son developed fever, malaise and diarrhoea
- Blood cultures grew *Salmonella* Typhi
- Treated with chloramphenicol with full recovery
Case

- Blood cultures from the mother, collected just prior to death also grew *Salmonella* Agona, resistant to ampicillin, chloramphenicol and gentamicin.
Case

- The husband and father worked in a hospital microbiology laboratory.
- He had subcultured stocks of both S. Typhi and S. Agona two weeks prior to the onset of his wife’s illness.
- Both of these isolates had been supplied as part of external QAP programs.
- Phage typing and antibiogram patterns were identical to the clinical isolates.
- He did not develop any illness, and stool cultures were negative.
- Cooked evening meals for the family.

J Clin Microbiol 13:855
Typhoid

- *Salmonella* Typhi and Paratyphi
- Accounts for more reported fatalities than any other laboratory acquired infection
- 50-100 cases of Typhoid fever reported in Australia annually
- Family members may acquire infection from laboratory staff
Typhoid

- Infection occurs from ingestion after contamination of hands
  - Hands may become contaminated by handling colonies or from contaminated surfaces or equipment
Typhoid

- Prevent laboratory exposure:
  - Suspect typhoidal salmonella:
    - Encourage requesting Drs to supply clinical information
    - Encourage laboratory staff to read clinical information (eg: ?fever in a returned traveller)
  - Use PPE
  - Hand hygiene
  - Vaccination
Other laboratory acquired infections

- **Bacteria**
  - *Shigella*, non-typhoidal *Salmonella*, *E. coli*, *Vibrio*
  - *Bacillus anthracis*
  - *Staphylococcus aureus*, *Streptococcus pyogenes*
  - *Burkholderia pseudomallei*, *Francisella tularensis*
  - Tuberculosis

- **Viruses**
  - HAV
  - HBV, HCV, HIV

- **Parasites**
  - Malaria
  - Strongyloides
Preventing laboratory acquired infection

- Laboratory containment requirements
- Communication
- Hand-hygiene
- Personal protective equipment
- Safety cabinets – proper use
- Education
- Vaccination
- TB Screening
Laboratory containment requirements

Australian/New Zealand Standard™
AS/NZS 2243.3:2010
Safety in laboratories
Part 3: Microbiological safety and containment
Laboratory containment requirements

(a) **Risk Group 1 (low individual and community risk)** — a microorganism that is unlikely to cause human or animal disease.

(b) **Risk Group 2 (moderate individual risk, limited community risk)** — a microorganism that is unlikely to be a significant risk to laboratory workers, the community, livestock, or the environment; laboratory exposures may cause infection, but effective treatment and preventive measures are available, and the risk of spread is limited.

(c) **Risk Group 3 (high individual risk, limited to moderate community risk)** — a microorganism that usually causes serious human or animal disease and may present a significant risk to laboratory workers. It could present a limited to moderate risk if spread in the community or the environment, but there are usually effective preventive measures or treatment available.

(d) **Risk Group 4 (high individual and community risk)** — a microorganism that usually produces life-threatening human or animal disease, represents a significant risk to laboratory workers and may be readily transmissible from one individual to another. Effective treatment and preventive measures are not usually available.
Communication

- Educate ordering clinicians to supply clinical notes
- Educate lab staff to read clinical notes
- If pathogen with potential to cause laboratory acquired infection is suspected: seal plates in parafilm
5 Moments for HAND HYGIENE

1. BEFORE TOUCHING A PATIENT
2. BEFORE A PROCEDURE
3. AFTER A PROCEDURE OR BODY FLUID EXPOSURE RISK
4. AFTER TOUCHING A PATIENT
5. AFTER TOUCHING A PATIENT’S SURROUNDINGS
0. Wet hands with water;
1. Apply enough soap to cover all hand surfaces;
2. Rub hands palm to palm;
3. Right palm over left dorsum with interlaced fingers and vice versa;
4. Palm to palm with fingers interlaced;
5. Backs of fingers to opposing palms with fingers interlocked;
6. Rotational rubbing of left thumb clasped in right palm and vice versa;
7. Rotational rubbing, backwards and forwards with clasped fingers of right hand in left palm and vice versa;
8. Rinse hands with water;
9. Dry hands thoroughly with a single use towel;
10. Use towel to turn off faucet;
11. Your hands are now safe.
PPE
Safety Cabinets
Education
Education
<table>
<thead>
<tr>
<th>Pathogen</th>
<th>Clinical details</th>
<th>Laboratory features</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Bacillus anthracis</strong></td>
<td>Raised skin lesion with black eschar from rural NSW</td>
<td>Non-haemolytic, large (3-5mm) white-grey, ground glass colonies with outgrowths – <em>Medusa head</em> colonies, large gram positive rod with, subterminal endospores</td>
</tr>
<tr>
<td><strong>Brucella sp</strong></td>
<td>Fever in returned traveller especially from the Middle East, India and Mediterranean PUO Pig-shooter</td>
<td>Very small gram negative coco-bacilli often in large clumps from BC bottle, slow growing, shiny grey-white growth – no discernable colonies at 24hrs incubation. Strict aerobe – only aerobic BC bottle will flag, (oxidase positive and catalase positive - but do not perform these tests if Brucella is suspected)</td>
</tr>
<tr>
<td><strong>Burkholderia pseudomallei</strong></td>
<td>Severe pneumonia or sepsis acquired in Northern Australia or Asia</td>
<td>Gram negative rod (&quot;Safety Pin&quot;), oxidase positive</td>
</tr>
<tr>
<td><strong>Dimorphic fungi</strong></td>
<td></td>
<td>Yeast from sterile site/BAL, slow growing (Histoplasma); mould from sterile site, ‘spherules’ in tissue on gram stain/histopathology (Coccidioides)</td>
</tr>
<tr>
<td><strong>Neisseria meningitidis</strong></td>
<td>Meningitis</td>
<td>Gram negative diplococci</td>
</tr>
<tr>
<td><strong>Salmonella sp.</strong></td>
<td>Diarrhoea</td>
<td>Mauve on salmonella chrome, pink with black centre on XLD, oxidase negative</td>
</tr>
<tr>
<td><strong>Salmonella Typhi or Paratyphi</strong></td>
<td>Fever in a returned traveller</td>
<td>Gram negative rod, clear on chromAgar</td>
</tr>
<tr>
<td><strong>Shigella sp.</strong></td>
<td>Diarrhoea</td>
<td>Clear on XLD, oxidase negative</td>
</tr>
<tr>
<td><strong>Vibrio sp.</strong></td>
<td>Diarrhoea</td>
<td>Green or Yellow on TCBS, usually curved gram negative rod, oxidase positive</td>
</tr>
</tbody>
</table>
## Vaccination for laboratory workers: CIDMLS

<table>
<thead>
<tr>
<th>Staffmember Duties</th>
<th>Special Vaccinations Needed*</th>
</tr>
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<tbody>
<tr>
<td>Processing or handling faeces specimens</td>
<td>Hepatitis A and <em>Salmonella</em> Typhi</td>
</tr>
<tr>
<td>Enteric Reference Laboratory</td>
<td><em>Salmonella</em> Typhi</td>
</tr>
<tr>
<td>Faeces plate reading</td>
<td><em>Salmonella</em> Typhi</td>
</tr>
<tr>
<td>24 &amp; 48 Hour plate reading</td>
<td><em>Neisseria meningitidis</em></td>
</tr>
<tr>
<td>Blood Culture, tissue bench plate reading</td>
<td><em>Neisseria meningitidis</em> and <em>Salmonella</em> Typhi</td>
</tr>
<tr>
<td>Identification Reference Laboratory</td>
<td><em>Neisseria meningitidis</em> and <em>Salmonella</em> Typhi</td>
</tr>
<tr>
<td>Arbovirus Reference Laboratory</td>
<td>Japanese Encephalitis, Yellow Fever, Rabies</td>
</tr>
<tr>
<td>P4 Facility</td>
<td><em>Neisseria meningitidis</em> and <em>Salmonella</em> Typhi</td>
</tr>
</tbody>
</table>

* These vaccinations are in addition to those required by NSW Health Policy directive: “Occupational Assessment, Screening and Vaccination Against Specified Infectious Diseases” (PD 2011_005)
It’s time for a centralized registry of laboratory-acquired infections

Karanjit Singh

A recent serious outbreak of Salmonella linked to clinical and teaching microbiology laboratories highlights the dangers of working with laboratory pathogens—but it is probably not an isolated occurrence. Without a better system for reporting infections resulting from laboratory exposures, we risk seeing more of these types of outbreaks.
Letters to the Editor

Sniffing Bacterial Cultures on Agar Plates: a Useful Tool or a Safety Hazard?

TABLE 1. Colony count from air sampled 2 cm from cultures on blood agar

<table>
<thead>
<tr>
<th>Organism</th>
<th>Colony count</th>
<th>No. of CFU/m³</th>
</tr>
</thead>
<tbody>
<tr>
<td>Neisseria meningitidis</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Streptococcus pneumonia</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Burkholderia pseudomallei</td>
<td>1</td>
<td>6.25</td>
</tr>
<tr>
<td>Bacillus spp.</td>
<td>1</td>
<td>6.25</td>
</tr>
<tr>
<td>Pseudomonas aeruginosa</td>
<td>1</td>
<td>6.25</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>2</td>
<td>12.5</td>
</tr>
<tr>
<td>Escherichia coli</td>
<td>1</td>
<td>6.25</td>
</tr>
<tr>
<td>Candida albicans</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Bacteroides fragilis</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

5000 sniffs in one m³
Our results suggest that the risk of inhaling a significant dose of a bacterium from solid agar is low. Despite this, we would caution staff against sniffing plate cultures that they suspect to include *Neisseria meningitidis*, *Brucella* spp., or other highly virulent pathogens.

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J Clin Microbiol 40:3877
Questions?