Standard Microbiological Practices

- Wash your hands often during work and before you leave.
- No outside activities like eating, drinking and smoking are permitted in lab areas.
- Mouth pipetting is not allowed.
- Follow handling procedures for sharps. Use engineering and work practice controls that reduce the risk of sharps injury.
- Minimize the creation of splashes and/or aerosols.
- Decontaminate surfaces after completion of work.
- A sign incorporating the universal biohazard symbol must be posted at the entrance to the laboratory when infectious agents are present. Posted information must include: the laboratory's biosafety level, the supervisor’s name, telephone number, and required procedures for entering and exiting the laboratory.
- Laboratory personnel must receive appropriate training annually or when procedural or policy changes occur.

Extra precautions to take with sharps are as follows:
- Carefully manage all needles and other sharps.
- Used disposable sharps must be placed in a puncture resistant container for disposal.
- Non disposable sharps must be placed in a puncture resistant container until disinfected.
- Broken glassware must not be handled directly.
- Plastic should be substituted for glass

Biological Waste:
Infectious waste must be handled according to EPA procedures as outlined in the Biosafety manual. Biological waste that does not meet the infectious waste definition should be treated according to lab specific procedures.

BSL -2
Biosafety Level 2

Biosafety level two builds upon BSL-1. BSL-2 is suitable for work involving agents that pose moderate hazards to personnel and the environment. It differs from BSL-1 in that: 1) laboratory personnel have specific training in handling pathogenic agents and are supervised by scientist competent in handling infectious agents and associated procedures; 2) access to laboratory is restricted when work is being conducted; 3) and all procedures in which infectious aerosols or splashes may be created are conducted in Biosafety cabinets (BSCs) or other physical contaminant equipment.

Detailed information about all of these topics can be found in the Ohio University Biosafety Manual. Information was gathered from the CDC and NIH.
Safety Precautions for Biosafety Level 2

Special Practices
- All persons entering the laboratory must be advised of the potential hazards and meet any necessary entry/exit requirements.
- The laboratory supervisors must ensure that laboratory personnel demonstrate proficiency in standard and special microbiological practice, before working with BSL-2 agents.
- Potentially infectious materials must be placed in a durable, leak proof container during collection, handling, processing, storage, or transport within a facility.
- Laboratory equipment should be routinely decontaminated, as well as, after spills and splashes, and before repair, maintenance, or removal.
- Animals and plants not associated with the work being performed must not be permitted in the laboratory.
- All procedures that may generate an infectious aerosol should be conducted within a BSC or other physical containment device.

Safety Equipment:
- Protective laboratory coats, gowns, smocks, or uniforms designated for laboratory use must be worn. Remove protective clothing before leaving for non-laboratory areas. It is recommended that laboratory clothing not be taken home.
- Eye and face protection is used for anticipated splashes or sprays of infectious or other hazardous materials when the microorganism must be handled outside the BCS or containment device. Eye and face protection must be disposed of with other contaminated laboratory waste or decontaminated before reuse. Persons who wear contact lenses should also wear eye protection.

Safety Equipment: Continued
- Gloves must be worn to protect hands from exposure to hazardous materials. Glove selection should be based on appropriate risk assessment. Alternatives to latex gloves should be available. In addition BSL-2 workers should:
  - Change gloves when contaminated, integrity has been compromised, or when otherwise necessary.
  - Remove gloves and wash hands when work with hazardous materials has been completed and before leaving the laboratory.
  - Do not wash or reuse disposable gloves. Dispose of used gloves with other contaminated laboratory waste.

Spills and Clean up:
Inside the BSC:
1. Leave the cabinet running; inform others of the spill.
2. You need disinfectant, absorbent and gloves.
3. Use a tool to remove sharps and put them in the correct waste container.
4. Spray all surfaces with disinfectant
5. If necessary flood surfaces with disinfectant
6. Wait the appropriate contact time.
7. Absorb up all spill material and disinfectant; then place absorbent materials into the appropriate waste container.
8. Inform the laboratory supervisor and EHS, if necessary.

Outside the BSC: (Less than 500 mL)
1. Inform others of the spill and get help if needed.
2. You need disinfectant, absorbent, gloves, eye goggles, and a lab coat.
3. Cover the spill with absorbent material.
4. Pour disinfectant over the spill and absorbent material.
5. Allow the appropriate contact time.

Outside the BSC cont.
6. Pick or scoop up absorbent material from the outside to the inside of the spill, and put directly into the appropriate waste container.
7. Rewipe the spill area with disinfectant.
8. Inform the laboratory supervisor and EHS, if necessary.

Outside the cabinet: (More than 500 mL)
Restrict access to the area and get help immediately. Contact laboratory supervisor and EHS.

Disinfectant Information:
(Detergents can be used prior to disinfectants, to break-up organic matter—esp. when using chlorine.)

<table>
<thead>
<tr>
<th>Chemical</th>
<th>Alchol</th>
<th>Contact Time</th>
<th>Inactivates?</th>
<th>Hazards</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chlorine Compounds (ex. Clorox)</td>
<td>10%</td>
<td>15 minutes</td>
<td>Bacteria, Most Viruses &amp; Some Spores</td>
<td>Flammable, Corrosive; Eye, Skin &amp; Respiratory Irritant; Toxic</td>
</tr>
<tr>
<td>Phenolic Compounds (ex. Lysol)</td>
<td>5%</td>
<td>10 minutes (or as listed on bottle)</td>
<td>Bacteria, Most Viruses (not effective on cox-sackie virus)</td>
<td>Corrosive; Skin &amp; Eye Irritant; Toxic</td>
</tr>
<tr>
<td>Iodophor (ex. Wesco-dyne)</td>
<td>1%</td>
<td>10 minutes (or as listed on bottle)</td>
<td>Bacteria, Most Viruses</td>
<td>Corrosive; Skin &amp; Eye Irritant; Toxic</td>
</tr>
<tr>
<td>Alcohols (isopropanol, ethanol)</td>
<td>70%</td>
<td>10 minutes</td>
<td>Bacteria, Most Viruses</td>
<td>Corrosive; Skin &amp; Eye Irritant; Toxic</td>
</tr>
</tbody>
</table>

Use Dilution (in water)
- 10%
- 5%
- 1%
- 70%

Contact Time
- 10 minutes
- 15 minutes

Inactivates?
- Bacteria, Most Viruses & Some Spores
- Bacteria, Most Viruses (not effective on cox-sackie virus)
- Bacteria, Most Viruses

Hazards
- Flammable
- Corrosive
- Eye, Skin & Respiratory Irritant
- Toxic