I. RECOGNITION OF BIOLOGICAL HAZARDS

A. Biohazard
   - Biohazardous materials and organisms include all infectious organisms (bacteria, chlamydia, fungi, parasites, rickettsias, viruses, etc.) that can cause disease in humans, or cause significant environmental or agricultural impact.
   - Other biohazards include work with human or primate tissues, fluids, cells or cell culture; recombinant DNA; transgenic plants or animals; human gene therapy; releases of recombinant DNA to the environment; and work with animals known to be reservoirs of zoonotic diseases.

II. EVALUATION OF BIOLOGICAL HAZARDS

B. Biosafety Level - The laboratory conditions under which the biohazardous agent can be safely handled.
   1. Levels
      - There are four biosafety levels. These levels have been summarized on the following table, which consist of combinations of laboratory practices and techniques, safety equipment, and laboratory facilities.
   2. Selection
      - The selection of an appropriate biosafety level is dependent upon a number of factors, most importantly the virulence, pathogenicity, biological stability and communicability of the agent, nature or function of the laboratory, quantity and concentration of the agent, endemicity of the agent, and availability of effective vaccines or therapeutic measures.
      - The principal investigator is primarily responsible for assessing risks and for implementing the recommended biosafety levels.
      - The biosafety level should be commensurate with that required for the agent of highest virulence known or likely to be encountered in the course of contemplated work. For example, all diagnostic sera of human origin (i.e., blood, body fluids, etc.) should be considered potentially infectious for Hepatitis and HIV.
      - If, in the course of diagnostic or other laboratory examinations there is evidence that the materials being studied contain an agent of higher than expected risk, the biosafety level should be raised accordingly.
### BIOLOGICAL HAZARDS

#### BMBL Section III – Summary of Recommended Biosafety Levels for Infectious Agents

<table>
<thead>
<tr>
<th>BSL</th>
<th>Agents</th>
<th>Practices</th>
<th>Safety Equipment (Primary Barriers)</th>
<th>Facilities (Secondary Barriers)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Not known to consistently cause disease in healthy adults</td>
<td>Standard Microbiological Practices</td>
<td>None Required</td>
<td>• Open bench top</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>• Sink required</td>
</tr>
</tbody>
</table>
| 2   | Associated with human disease, hazard = percutaneous injury, ingestion, mucous membrane exposure | BSL-1 practices plus:  
- Limited access  
- Biohazard Warning signs  
- "Sharps" precautions  
- Biosafety manual defining any needed waste decontamination or medical surveillance policies | Primary barriers=Class I or II BSCs or other physical containment devices used for all manipulations of agents that cause splashes or aerosols of infectious materials; PPEs: laboratory coats, gloves, face protection as needed | BSL-1 plus:  
- Autoclave available |
| 3   | Indigenous or exotic agents with potential for aerosol transmission; disease may have serious or lethal consequences | BSL-2 practices plus:  
- Controlled access  
- Decontamination of all waste  
- Decontamination of lab clothing before laundering  
- Baseline serum | Primary barriers=Class I or II BSCs or other physical containment devices used for all open manipulations of agents; PPEs: protective lab clothing; gloves; respiratory protection as needed | BSL-2 plus:  
- Physical separation from access corridors  
- Self-closing, double-door access  
- Exhausted air not recirculated  
- Negative airflow into laboratory |
| 4   | Dangerous/exotic agents which pose high risk of life-threatening disease, aerosol-transmitted lab infections; or related agents with unknown risk of transmission | BSL-3 practices plus:  
- Clothing change before entering  
- Shower on exit  
- All material decontaminated on exit from facility | Primary barriers=All procedures conducted in Class III BSCs or Class I or II BSCs in combination with full-body, air-supplied, positive pressure personnel suit | BSL-3 plus:  
- Separate building or isolated zone  
- Dedicated supply and exhaust, vacuum and decon systems  
- Other requirements outlined in the text |

#### BMBL Section IV – Summary of Recommended Biosafety Levels for Activities in Which Experimentally or Naturally Infected Vertebrate Animals Are Used

<table>
<thead>
<tr>
<th>BSL</th>
<th>Agents</th>
<th>Practices</th>
<th>Safety Equipment (Primary Barriers)</th>
<th>Facilities (Secondary Barriers)</th>
</tr>
</thead>
</table>
| 1   | Not known to consistently cause disease in healthy human adults        | Standard animal care and management practices, including appropriate medical surveillance programs                  | As required for normal care of each species                                                                                         | • Standard animal facility  
- No recirculation of exhaust air  
- Directional air flow recommended  
- Hand washing sink recommended |
| 2   | Associated with human disease. Hazard: percutaneous exposure, ingestion, mucous membrane exposure | ABSL-1 practices plus:  
- Limited access  
- Biohazard warning signs  
- Sharps precautions  
- Biosafety Manual  
- Decontamination of all infectious wastes and of animal cages prior to washing | ABSL-1 equipment plus primary barriers: containment equipment appropriate for animal species; PPEs: laboratory coats, gloves, face and respiratory protection as needed | ABSL-1 facility plus:  
- Autoclave available  
- Hand washing sink available in the animal room  
- Mechanical cage washer used |
| 3   | Indigenous/exotic agents with potential for aerosol transmission; disease may have serious health effects | ABSL-2 practices plus:  
- Controlled access  
- Decontamination of clothing before laundering  
- Cages decontaminated before bedding removed  
- Disinfectant foot bath as needed | ABSL-2 equipment plus:  
- Containment equipment for housing animals and cage dumping activities  
- Class I or II BSCs available for manipulative procedures (inoculation, necropsy) that may create infectious aerosols; PPEs: appropriate respiratory protection | ABSL-2 facility plus:  
- Physical separation from access corridors  
- Self-closing, double-door access  
- Sealed penetrations  
- Sealed windows  
- Autoclave available in facility |
| 4   | Dangerous/exotic agents that pose high risk of life threatening disease; aerosol transmission, or related agents with unknown risk of transmission | ABSL-3 practices plus:  
- Entrance through change room where personal clothing is removed and laboratory clothing is put on; shower on exiting  
- All wastes are decontaminated before removal from the facility | ABSL-3 equipment plus:  
- Maximum containment equipment: Class III BSCs or Class I or II BSCs in combination with full-body, air-supplied, positive pressure personnel suit used for all procedures and activities | ABSL-3 facility plus:  
- Separate building or isolated zone  
- Dedicated supply and exhaust, vacuum and decon systems  
- Other requirements outlined in the text |

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2 U.S. Department of Health and Human Services Centers for Disease Control and Prevention and National Institutes of Health, “Biosafety in Microbiological and Biomedical Laboratories”,  
B. Waste

1. Biohazardous Waste - Biohazardous waste means any of the following:
   - Human or animal specimen cultures from medical and pathological laboratories, cultures and stocks of infectious agents from research laboratories, waste from the production of bacteria, viruses or the use of spores, discarded live and attenuated vaccines, culture dishes and contaminated devices used to transfer, inoculate, and mix cultures.
   - Human surgery specimens or tissues removed at surgery or autopsy, which are suspected by the attending physician or dentist of being contaminated with infectious agents’ known to be contagious to humans.
   - Animal parts, tissues, fluids, or carcasses suspected of being contaminated with infectious agents’ known to be contagious to humans.
   - Waste which contains recognizable fluid blood, fluid blood products, containers, or equipment containing fluid blood or blood from animals, having been infected with diseases that are highly communicable to humans.
   - Waste containing discarded materials contaminated with excretion, exudates, or secretions from humans who are required to be isolated to protect others from communicable diseases or isolated animals having been infected with diseases communicable to humans.
   - Waste which is hazardous only because it is comprised of human surgery specimens or tissues which have been fixed in formaldehyde or other fixatives, or only because the waste is contaminated through contact with, or having previously contained chemotherapeutic agents, including, but not limited to, gloves, disposable gowns, towels, and intravenous solution bags and attached tubing which are empty. (Chemotherapeutic agent means an agent that kills or prevents the reproduction of malignant cells.)
   - Waste that is hazardous only because it is comprised of pharmaceuticals.

2. Medical Waste - Medical waste is laboratory, pathology or sharps biohazardous waste which is generated or produced as a result of:
   - diagnosis, treatment, or immunization of human beings or animals
   - research
   - producing or testing biologicals. (Biologicals mean medicinal preparations made from living organisms and their products, including, but not limited to, serums, vaccines, antigens, and antitoxins.) that has been infected with potential or known diseases communicable to humans.

3. Sharps Waste - Any device having acute rigid corners or edges, or projections capable of cutting or piercing, including hypodermic needles, syringes, blades, needles, broken glass items, pipettes and vials which are contaminated with other medical waste.
III. CONTROL OF BIOLOGICAL HAZARDS

A. Working with Human Biohazards - Follow the recommended biosafety level practices and procedures for the agent(s) used in the lab. Some key practices to be followed when working with human biohazards:
   - A hazard warning sign incorporating the biohazard symbol must be posted on access doors and on equipment where human biohazards are used or stored.
   - Use personal protective equipment (PPE) such as gloves, lab coat, etc., when handling human biohazards.
   - Use a biosafety cabinet when handling human biohazards, particularly when procedures may generate aerosols or splashing.
   - Decontaminate all work surfaces after completion of work.
   - Properly dispose of all waste generated from working with human biohazards (i.e., Accumulation Site, chemical disinfections, or disposal through an approved vendor.
   - Contact EH&S @ 530-752-1493

B. Handling “Other” Medical and Sharps Waste
   - Waste, which is contaminated through contact with, or having previously contained, chemotherapeutic agents, shall be segregated for storage. This type of waste must be placed in a secondary container, which shall be labeled on the lid and the sides with the words "Chemotherapy Waste", "CHEMO", or other labels approved EH&S. The label must be visible from any lateral direction, to ensure treatment of the biohazardous waste. Chemotherapy waste is picked-up by a vendor for final treatment at an off-site facility. (Contact EH&S @ 530-752-1493 additional information.)
   - Biohazardous waste which is comprised of human surgery specimens or tissues which have been fixed with formaldehyde or other fixatives, shall be segregated for storage then disposed of by incineration at an off-site facility. (Contact EH&S @ 530-752-1493 additional information.)
   - Liquid or semi-liquid biohazardous waste, such as blood or culture solutions, must be properly decontaminated prior to being discharged into the public sewage system. Example of proper decontamination of liquid waste is adding bleach solution and allowing at least 30 minutes of contact time.

1. “Red Bags”
   - Non-sharp infectious waste must be placed in red biohazard bags that are labeled with the words “Biohazardous Waste” or with the international biohazard symbol and the word “Biohazard.” During accumulation, red bags must be placed in a rigid secondary container.
   - Full bags should be tied to prevent leakage or expulsion of contents during future storage, handling or transport. (Recommendation: Bags should not be more than 2/3 full and use tape to seal bag.)
   - During storage, full untreated, red bags must be placed in a rigid container which are leak resistant, have tight fitting covers and kept clean and in good repair. Containers may be any color and labeled on the lid and on the sides with the words "Biohazardous Waste" or with the international
BIOLOGICAL HAZARDS

biohazard symbol and the word "BIOHAZARD" so as to be visible from any lateral direction.

- Red bags must be treated by a campus approved method (e.g. by UCDMC or a vendor for final treatment at an off-site facility. (Contact EH&S @ 530-752-1493 for additional information.)
- Full biohazard bags shall not be stored above 0°C (32°F) for more than seven days or below 0°C (32 °F) for more than 90 days before treatment.

2. Sharps Containers

- Place all sharps waste into appropriate sharps container. Sharps containers may be purchased through UCD Central Storehouse. (Order through UCD the Central Storehouse on-line: http://materiel.ucdavis.edu/storehouse/
- Do not overfill sharps containers. Do not shake up the container to try to fit more materials into it. Shaking the container aerosolizes the materials contained within.
- Sharps containers are “Single Use Only”. Do not reuse sharps containers.
- Sharps containers ready for disposal must be tightly sealed or taped to ensure that contents will not spill. Full sharps containers should not be stored longer than 7 days. Place “capped” sharps containers in the Medical Waste Toter at your Medical Waste accumulation site.

C. Biosafety Cabinets - Biosafety cabinets are designed to protect you from splashes and aerosols that are contaminated with biohazards.

1. Airflow

- Place necessary materials in the biosafety cabinet before beginning work to minimize the number of arm movement disruptions across the fragile air barrier of the cabinet.
- Ensure that the front grille of the cabinet is not blocked with any materials (i.e., absorbent paper, notebook, etc.) or equipment to allow cabinet to function properly.
- Place all materials as far back as practical, toward the rear edge of the work surface and away from the front grille of the cabinet to take advantage of the air split in the center of the cabinet.
- Delay manipulation of materials for approximately one minute after placing hands/arms inside the cabinet to allow stabilization of air in the cabinet.

2. Control Methods for Contamination

- Disinfect the work surface, interior walls, and interior surface of window of the cabinet to reduce contamination of materials to be used in the cabinet.
- Disinfect surfaces of materials and containers placed into the cabinet to minimize contamination of cultures.
- Do not bring potentially contaminated materials out of the cabinet until they have been surface decontaminated or placed in a closeable container for proper decontamination.
- Decontaminate surfaces of all containers and equipment removed from the cabinet when work is completed.
- Wipe down the cabinet's work surface, sides, back, and interior of the glass at the end of the workday.
• Decontaminate biosafety cabinets before HEPA filters are changed or internal work is done and before cabinet is relocated. The most common method for this type of decontamination is the use of formaldehyde gas. An EH&S approved vendor must conduct this decontamination procedure.

3. Work practices
• Turn cabinet on for approximately three to five minutes to allow it to purge or remove any particulates in the cabinet.
• Wear personal protective equipment such as lab coat, gloves, etc., to protect the worker from contact with biohazardous materials used in the cabinet.
• Adjust stool/seat height so that worker's face is above the front opening of the cabinet.
• Place plastic-backed absorbent paper on the work surface (note: ensure that grilles are not blocked by the absorbent paper), if desired. This facilitates routine cleanup and reduces splatter and aerosol formation during a spill. Absorbent toweling must be properly decontaminated prior to disposal.
• Arrange materials within the cabinet to allow active work to flow from the clean to contaminated area across the work surface. (Limit the movement of "dirty" items over "clean" items.) This reduces the potential for cross-contamination in the cabinet.
• Place bulky items such as biohazard bags, discard pipette trays, and suction collection flasks to one side of the interior of the cabinet to minimize risk of cross-contamination.
• Do not tape a biohazard collection bag to the outside of the cabinet. Do not use upright pipette collection containers in the cabinet or place them on the floor outside the cabinet. Frequent inward and outward movement needed to place objects in these containers is disruptive to the integrity of the cabinet air barrier and can compromise personnel and product protection.
• Follow good microbiological techniques when working in a cabinet. For example, techniques to reduce splatter and aerosol generation will minimize the potential for personnel exposure to infectious materials manipulated within the cabinet.
• Do not use open flames in a cabinet. An open flame in a biosafety cabinet creates turbulence that disrupts the pattern of air supplied to the work surface. Open flames are not required in the near microbe-free environment of a biosafety cabinet. If absolutely necessary, touch-plate micro burners equipped with a pilot light to provide a flame on demand may be used. (Internal cabinet air disturbance and heat buildup will be minimized.)
4. Certification

All campus biosafety cabinets must be certified annually, to ensure that unit is functioning properly. Contact Technical Safety Service (TSS) @ 800-877-7742 to conduct biosafety cabinet certification.

Source(s):
Biosafety in Microbiological and Biomedical Laboratories , CDC, NIH, U.S. Dept. of Health and Human Resources, 4th edition, May 1999