Biosafety

The following experiments should be registered with the applicable committee(s) and safety personnel as necessary:

- Recombinant DNA (rDNA) activities
- Work with potentially infectious agents and biological toxins
- Experiments involving the use of potentially infectious human and/or non-human primate materials, such as unfixed tissues, cell lines, and body fluids/excretions/secretions.

The Institutional Biosafety Committee (IBC):

It is the duty of the IBC to develop policies and procedures relating to pathogenic microorganisms, biological toxins, rDNA and human and non-human primate materials and implement biological safety programs. For more information on the IBC, contact UCA's Department of Biology.

BIOSAFETY LEVELS (BSL)

General safety considerations, as outlined in the Chemical Hygiene Plan, should be followed for biological and biochemistry labs in the Department of Chemistry. Additional safety measures are determined based on the BSL of each individual lab. Four levels of biosafety are defined in the BMBL http://www.cdc.gov/biosafety/publications/index.htm. The levels build upon each other in ascending order by degree of protection provided to personnel, the environment, and the community. This means that the requirements for a higher BSL also includes all safety measures from each lower BSL. Each level consists of combinations of laboratory practices, safety equipment, and laboratory facilities which allow manipulation of biological agents of increasing danger to life and health.

- BSL1 – BSL1 laboratory facilities and practices are suitable for work involving well characterized agents not known to consistently cause disease in immunocompetent adult humans. Examples include B. subtilis, E. coli, and L. acidophilus.
- BSL2 – BSL2 laboratory facilities and practices are suitable for work involving agents that pose a moderate hazard to personnel and the environment. Treatment for disease is generally available; however, illness is sometimes fatal. Examples include Salmonellae, Hepatitis B virus, bloodborne pathogens, and human body fluids, particularly when visibly contaminated with blood.
- BSL3 – BSL3 laboratory facilities and practices are suitable for work with indigenous or exotic agents that may cause serious or potentially lethal disease through inhalation as they present the potential for aerosol transmission. Treatment for exposure or vaccines may be available. Specific training, biological safety cabinet use, and special engineering and design features are required. Training and setup for BSL3 laboratories will have to cooperate with the IBC and, potentially, the CDC as these agents are largely part of the select CDC programs. Examples include H5N1 Influenza virus, Bacillus anthracis, Yersinia pestis, Burkholderia, Francisella tularensis, Brucella, Clostridium botulinum, Mycobacterium tuberculosis, Coxiella burnetii, Hantavirus, and West Nile virus.
- BSL4 – Organisms are highly pathogenic and require handling in special laboratory facilities designed to contain them. Examples include viral hemorrhagic fevers, such as Ebola, Lassa fever, Hantavirus pulmonary syndrome (HPS) and hemorrhagic fever with renal syndrome (HFRS).

Recommended PPE/Equipment

- BSL1 – Standard PPE, no special equipment
- BSL2 – Standard PPE, biological safety cabinets or other containment devices for agents that likely aerosolize or cause splashes, autoclave
- BSL3 and BSL4 – The UCA Department of Chemistry currently does not have any facilities or laboratories approved for BSL3 or BSL4 work

Biological Spill Kit

A biological spill kit shall should contain items in a normal spill kit (absorbent material, hand broom, dust pan, and a pair of sturdy rubber gloves) plus appropriate disinfectant, tongs or other grasping tool(s), and an autoclavable secondary container if an autocalve is accessible to the lab. Hand brooms and dust pans should only be used to sweep up spills that do not involve biohazards. Broken glass contaminated with a biohazard should be picked up with tongs and placed in the secondary container to be autoclaved.

General Spill Cleanup Guidelines for Biological Spill Response

Each laboratory has a unique combination of hazards and thus requires customized laboratory procedures for controlling spills. Below are guidelines for developing such procedures for managing biological spills in the laboratory. Do not hesitate to contact appropriate personnel if you are not comfortable addressing a spill.

- Alert others working in the area of the spill to prevent spreading. Post a sign if needed.
- If your lab coat and gloves are contaminated in the spill, promptly remove them by turning the exposed surface to the inside, placing them in a biohazard bag, and then thoroughly washing any affected areas with soap and water. You should remove your lab coat before removing your gloves.
- Evacuate the immediate area (10 foot radius) of the spill for a minimum of 20 minutes. In some laboratories this may be the entire laboratory space.
- Don clean PPE before addressing the spill. Wear at least gloves and safety glasses. A lab coat is recommended.
- Cover spilled material with paper towels. For large spills, use pig pads and/or absorbent dams. Do not neglect furniture, equipment and vertical surfaces (cabinets, walls, doors). If commercial biological fluid solidifiers are used, follow the manufacturer's instructions.
- Starting at the perimeter, cover absorbent material with fresh disinfectant in sufficient quantity to ensure effective microbial inactivation. Be sure to use an appropriate disinfectant for the spilled material.

- Allow 20 minutes of contact time for disinfection unless a longer contact time is required by the manufacturer. Leave the immediate area while waiting for contact time to pass. Safely remove PPE and wash hands before leaving the laboratory.
- Don fresh gloves. The same lab coat may be worn if not contaminated in the previous step. Dispose of absorbent materials in biohazard waste container to be autoclaved unless bleach is used.
- Disinfect spill area again with diluted disinfectant and allow adequate contact time. Do not neglect vertical surfaces. It is prudent to mop the lab floor.
- Remove lab coat. Autoclave lab coats or soak in bleach to disinfect. Remove gloves and place in biowaste.
- Wash hands with soap and water when finished. Don clean PPE before continuing work.
- If broken glass is involved in a spill, remove visible pieces prior to covering the spill. Always handle glass indirectly using forceps, tongs or other device. Place glass in rigid container and decontaminate by autoclaving or chemical disinfection. Once disinfected, transfer glass to broken glass box – again, do not handle glass with your hands.

Spills in a Centrifuge

- Close centrifuge lid and let sit for 20 minutes to allow aerosols within the centrifuge to settle. While you are waiting, disinfect the exterior of the centrifuge by saturating with disinfectant soaked paper towels and allowing appropriate contact time. Follow with water or 70% ethanol/isopropanol if bleach was used.
- Open the centrifuge and carefully remove any pieces of debris from the centrifuge interior using forceps and place in a biowaste bag. Remove rotor and set aside for disinfection. It would be prudent to disinfect the rotor in a biological safety cabinet (BSC).
- Absorb/cover spill with paper towels. Squirt disinfectant on towels and remaining interior of centrifuge using a laboratory soak bottle or pour disinfectant. Let disinfectant sit for 20 minutes. Do not use a spray bottle as this can aerosolize contaminants.
- Follow with DI-water if bleach was used, then 70% ethanol or isopropanol. Dispose of all waste as biohazardous waste.

Potential Cleaning Agents

- **Sodium Hypochlorite (bleach)**
  - Recommended dilution is 1:5 to 1:100 in water (20% to 1% dilution).
  - Contact time varies with agent to be neutralized and concentration of solution.
  - Bleach solutions are effective against vegetative bacteria, fungi, and most viruses at 1:100 dilution.
  - Minimum 1:10 dilution is recommended for BSL2 activities; 1:5 dilution is needed to inactivate Mycobacterium.
  - Available free chlorine is maximized when the solution pH is 5-7.
  - Prepared solutions should be stored in brown plastic bottles to prevent decomposition under light.
  - Strips to test free chlorine ppm can be purchased.

        - ❖ Bleach solutions can be highly corrosive.
        - ❖ Bleach solutions work best when recently made, i.e. daily.
        - ❖ Bleach mixed with acids can release chlorine gas.
        - ❖ Bleach mixed with ammonia can release chloramine vapors.
- **Alcohols (ethanol, isopropanol)**
    - The effective dilution for decontamination is 60-80%; 70% is ideal
    - Effective against a broad spectrum of bacteria and many viruses
    - Ethanol is preferred to isopropanol given it has a slightly more broad-spectrum kill. Ethanol inactivates all lipophilic viruses and many hydrophilic viruses. Isopropanol is not active against hydrophilic viruses but virucidal against lipophilic viruses.
    - Alcohol waste from submersion must be disposed of as chemical waste
        - ❖ Not effective against bacterial spores, C. difficile, Helicobacter, and protein-rich materials such as blood or plasma.
        - ❖ Evaporates rapidly which reduces exposure time.
        - ❖ Can coagulate proteins and attach them to surfaces.
        - ❖ Can dissolve some plastics, rubbers, and adhesives.
- **Hydrogen Peroxide (3-8%)**
    - Surface sterilizer.
    - Effective across a broad spectrum of biohazards.
    - Rapidly oxidizes organic material.
        - ❖ Higher concentrations can be corrosive to skin and various metals.
        - ❖ Rapid oxidization of organic material means peroxide is consumed quickly and limits cleaning power.