

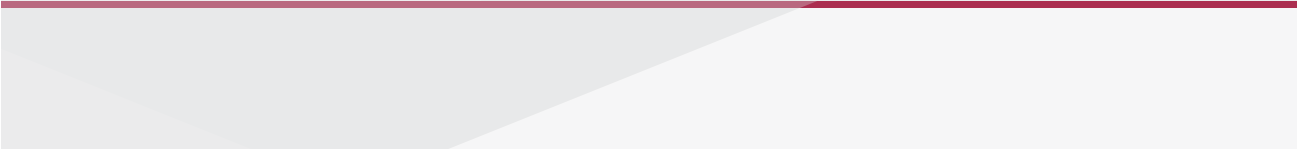
# BIOSAFETY MANUAL

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**USC** University of  
Southern California

Office of  
Environmental  
Health and Safety



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## 1.0 Purpose and Scope

The USC Biosafety Manual provides health and safety information to all USC employees and students who handle potentially hazardous biological materials (e.g., rDNA, animals, and biohazardous agents) and assists in minimizing or eliminating associated risks. It outlines the requirements of the USC Biosafety Program, including guidance and direction on how to complete a Biohazard Use Authorization (BUA) for all research involving biohazards and serves as a general reference source for the USC community.

The USC Biosafety Program covers any individual who works, attends, or volunteers at USC and handles potentially hazardous biological materials.

The Biosafety Program serves the community by:

- Providing information on safe procedures and practices.
- Explaining the proper use of safety equipment.
- Matching personal protective equipment to the task.
- Providing access to medical evaluations based on one's potential exposure to biohazardous materials.



The USC research community's efforts in continuing high safety standards within its laboratories encourage a strong safety culture; support employee health and well-being; limit or remove disruptions in research and productivity due to injury and illness; and, secure a competitive edge.

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## 2.0 Regulatory Requirements

Employee safety is regulated at the Federal level by the Occupational Safety and Health Administration (OSHA); however, states are permitted to regulate employee safety themselves, provided state regulation is at least as strict as the Federal standards. In California, employee safety is regulated by the Division of Occupational Safety and Health (DOSH), which is more commonly called Cal-OSHA or Cal/OSHA. Cal-OSHA regulations are found in Title 8 of the California Code of Regulations (CCR).

The USC Biosafety Program requires that the handling of all biological materials at USC follow the safety principles outlined in this manual, all related university policies and programs, and applicable state and federal regulations and guidelines.

The USC Biosafety Manual outlines the requirements and procedures developed by the USC Biosafety Program and incorporates applicable university safety policies and programs into its scope. Environmental Health & Safety (EH&S) Biosafety advises the reader/user to consult the following authoritative sources for mandated requirements and supplemental information in specific safety areas.

- [Chemical Hygiene Plan](#) covers chemical safety and many areas of general laboratory safety. It is intended to be a useful resource for USC laboratory personnel and principal investigators (PI), providing accessible and relevant information to enable safe operations.
- [Radiation Safety Manual/Program](#) offers procedural guidelines and information for prudent work practices while using any radioactive materials or devices at USC. The manual is made available to every area authorized to use radioactive materials and any area where radiation-producing machines are present.
- [Laser Safety Manual/Program](#) delineates safe work practices (engineering/administrative controls and PPE) for all faculty, staff, students, and volunteers who handle lasers at USC labs/locations. Additionally, it provides requirements for: (a) all non-clinical uses of Class 3B or Class 4 lasers and (b) anyone handling (or in the vicinity of) Class 3B and 4 lasers.
- [Personal Protective Equipment \(PPE\) Standard](#) mandates minimum PPE requirements for students, employees, and visitors entering areas with known or potential exposure(s) to hazardous materials (i.e., chemicals, biological agents, and radiological materials) and dangerous equipment.

- **[USC Injury and Illness Prevention Policy](#)** outlines safety responsibilities and requirements placed on all USC personnel (faculty, staff, students, contractors, and volunteers)—to support injury and illness prevention, maintain a safe and healthful workplace, and ensure individual and institutional compliance with relevant environmental health and safety regulations. This policy is part of the university’s Injury and Illness Prevention Program (IIPP), as required by California Title 8, General Industry Safety Orders, Section 3203.

### **State of California Regulations**

[The Bloodborne Pathogens Standard \(8CCR Sec. 5193\)](#), CalOSHA

[The Aerosol Transmissible Diseases Standard \(8CCR Sec. 5199\)](#), CalOSHA

[Medical Waste Management Act of California, January 2017](#), CDPH-EMB

### **Federal Regulations and Guidelines**

[The NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules](#), NIH OSP

[Biosafety in Microbiological and Biomedical Laboratories \(BMBL\)](#), NIH/CDC

[Federal Select Agent Program](#) (FSAP)

[Dual Use Research of Concern](#) (DURC)

[Department of Transportation](#) (DOT)



## 3.0 Roles and Responsibilities

### Institutional Biosafety Committee (IBC)

The USC IBC is responsible for creating and enforcing USC policies, practices, and guidelines related to the use of potentially hazardous biological agents such as infectious agents, human and non-human primate materials (including established cell lines), toxins of biological origin, regulated carcinogens, chemotherapy drugs, chemicals used in biomedical research, select agents, recombinant and synthetic nucleic acid molecules, and studies involving human gene transfer or the use of biohazardous materials in humans. The IBC has published a policy regarding IBC review and approval on the [IBC webpage](#). The full version of the USC IBC charge is available at <https://ehs.usc.edu/files/IBC-Charge-2017.pdf>.

The USC IBC is responsible for ensuring that research conducted using any of the above-mentioned agents does not endanger the researcher, laboratory workers, human research subjects, the public, or the environment. **It is the responsibility of the IBC to:**

- Formulate and implement policies related to the safe use of biological materials and some chemicals commonly used in biomedical research;
- Review research protocols involving biohazards and chemical hazards commonly used in biomedical research;
- Approve or disapprove such projects based on their potential hazard and proposed containment procedures;
- Establish, approve and monitor proper laboratory conditions and procedures required for such projects;
- Review the qualifications and training of investigators and laboratory personnel engaged in such research to ensure that appropriate laboratory safety techniques are used;
- Ensure adoption of proper disposal and decontamination procedures;
- Adopt emergency plans that cover accidental spills and personnel contamination resulting from research; and
- Ensure investigation and reporting of any significant problems with or violations of the NIH and CDC Guidelines, as specified in the *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules* and in the *CDC/NIH Biosafety in Microbiological and Biomedical Laboratories*.

The USC IBC meets monthly, usually on the fourth Thursday of the month. Except for November and December which have amended schedules for the holidays, the committee accepts BUAs for review through the first Monday of each month. A quorum for the IBC is the presence of at least 50% of the members. Members' presence may be in person or by telephonic or videographic means.

Protocols are approved by a simple majority. The USC IBC reports administratively to the President through

the Senior Vice President for Administration, and works closely with the Vice President for Research.

## Office of Environmental Health & Safety

The Office of Environmental Health and Safety is responsible for managing and implementing the USC Biosafety Program. The Biosafety Program consists of a Biosafety Officer (BSO) and Biosafety Specialists (BSPs).

### Biosafety Officer (BSO) and Biosafety Specialists (BSPs)

The BSO is a member of the IBC and reports to the Deputy Director, EH&S. The BSO is responsible for managing the Biosafety Program and ensuring that research with biological hazards is conducted safely and in full compliance with all applicable regulations.

The BSO is the primary subject matter expert on biosafety-related items and is consulted when risk assessments are required to determine how to safely work with biological hazards. The BSO supervises the Biosafety Specialists who perform many of the functions associated with the program.

The BSO and BSPs:

- Review the Biohazardous Use Authorizations (BUAs) applications electronically submitted by Principal Investigators (PIs).
- Evaluate each BUA for completion and accuracy prior to submitting it for review by IBC members.
- Oversee the review and approval process by the IBC.
- Develop general policies for biosafety.
- Develop and distribute information relevant to biosafety.

- Evaluate:
  - Equipment and physical facilities associated with projects involving biohazardous materials.
  - Operational techniques and procedures.
- Conduct:
  - Risk assessments when required.
  - Training programs.
  - Inspections of laboratory facilities that use biological materials.
- Assure that laboratory procedures are in accordance with USC policies and applicable biosafety regulations.
- Provide advice on the decontamination of facilities and equipment following spills or prior to remodeling or modification of facilities.
- Respond to emergencies and investigate accidental biological exposures.
- Coordinate with IACUC regarding approval of biohazardous materials in animal research.
- Coordinate with IRB regarding approval of the use of biohazards in clinical trials.
- Assure compliance with the Federal Select Agent Program and the Dual Use Research of Concern policy.

## Principal Investigator (PI)

The PI is ultimately responsible for all aspects of safety in his or her laboratory including the safe handling of biohazardous material. The PI must comply with and assure that the researchers listed on the BUA comply with all applicable regulations. EH&S staff, specifically the BSO and Biosafety Specialists, provide guidance to the PI on the requisite regulations.

It is the responsibility of the PI to:

- Ensure a safe working environment.
- Provide instructions and training on safe and proper biosafety practices to all persons working within their research area.
- Provide emergency procedures for laboratory personnel. These procedures must include the names and telephone numbers of key lab personnel (e.g., Lab Manager, Safety Officer, other) to be contacted in case of emergency. These procedures will be prominently posted in work areas where biohazardous materials are used.
- Maintain adequate control of the biohazardous materials.
- Provide necessary personal protective equipment (PPE) for safe work with hazardous materials.
- Properly label all areas where biohazards are stored or handled.
- Notify the BSO or Biosafety Specialists of any accident or abnormal incident involving or suspected of involving biohazardous material.
- Inform the IBC of:
  - Changes in personnel and any significant changes in lab design or procedures by updating the BUA.

- Plans to relocate the lab or leave the university via the BSO or Biosafety Specialists.
- Ensure that researchers complete initial and annual refresher biosafety training.
- Provide easy access to Safety Data Sheets (SDSs).
- Prepare Standard Operating Procedures (SOPs) including safe work practices for all routine processes involving biohazardous materials conducted in the facility.
- Hold regular safety meetings.
- Determine and document the personal protective equipment (PPE) needed for each procedure.

### Research Staff

The research staff members are individuals who work under the supervision of the PI and conduct research with biohazardous materials in an area for which the PI is responsible. Research staff members have the responsibility to practice safe use and handling of biohazardous materials.

Other responsibilities include:

- Read, understand, and follow university safety policies and standards as well as guidelines outlined in the USC Biosafety Manual.
- Attend appropriate EH&S biosafety training.
- Attend site-specific safety training by PI or lab manager.
- Follow SOPs and safe handling practices when using biohazardous materials.
- Understand risks associated with their research and ask questions for any items that are unclear to them.

- Know where the nearest eyewash/safety shower stations and fire extinguishers are and the emergency evacuation route for the building.
- Follow proper emergency procedures if an accident or incident occurs while handling biohazardous materials.
- Report any unsafe practices or concerns to the BSO or Biosafety Specialists.
- Notify the BSO or Biosafety Specialists regarding any accident or incident that occurs with infectious agents, chemicals, or rDNA while performing biomedical research.
- Refer to the [Recombinant DNA and Biohazard Incident Reporting Fact Sheet](#) (see figure 3.1) for additional information.

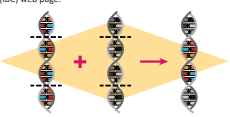
**Figure 3.1. Reporting Biohazard Incidents**

FactSheet

Recombinant DNA and  
Biohazard Incident Reporting

**P**ersonnel who work with recombinant DNA (rDNA) are required to report significant spills, incidents, potential or accidental exposures, or illnesses associated with rDNA research and other biohazardous agents when they occur. This is not only a regulatory requirement, but protection for coworkers and the community at large.

All potential injuries or exposures to biohazardous materials (including recombinant DNA) must be reported using the steps listed in the exposure control plan for your laboratory. If you don't have an exposure control plan you can find one at the [USC Institutional Biosafety Committee \(IBSC\)](#) web page.



Take care of the injury or exposure to yourself **FIRST**. Whether you were stuck with a needle or a sharp; splashed in the eyes, mouth or nose; or have experienced an unusual reaction to something you are working with, the area affected must be washed or rinsed first and the incident reported to your supervisor. Medical attention may be necessary.

If the incident involves recombinant DNA (rDNA), additional reporting is necessary. NIH has specified certain types of incidents for reporting.

These include the following:

- Overt exposure to recombinant organisms.
- Injuries or illnesses stemming from a potential biohazard exposure to recombinant organisms.
- Non-injury incidents, spills and exposures while using recombinant DNA.
- Release or escape of any transgenic organisms (e.g. transgenic flies, animals, and plants) from containment.
- Inadvertent or accidental spills of rDNA outside of containment.

**WHAT I NEED TO KNOW**

- Ensure your lab has an exposure control plan.
- Treat any exposure or injury immediately.
- Report all exposures and injuries when they occur.
- Report all non-injury incidents, spills and exposures while using recombinant DNA to EH&S Biosafety.
- Post emergency numbers and contacts in the laboratory and review with all personnel.

**Spills, accidents, and incidents (even minor ones) must be reported.** If you have questions about reporting a potential exposure, how to clean up a spill, or how to decontaminate a particular biohazard, EH&S and Biosafety personnel are available to help: 323-442-2200 or [biosafety@usc.edu](mailto:biosafety@usc.edu).


Call the Department of Public Safety (DPS) at 323-442-1000 (HSC) or 213-740-4321 (UPC) to report an emergency. DPS will assist with additional support from On-Call staff. Serious injuries or illnesses must be reported through the Injury Notification hotline: 323-442-2200, then press 1. Provide detailed information e.g., name of injured party; time of injury; location; witnesses; etc.

Note that a [Manager's Report of Incident](#) must be completed and submitted to Workers Compensation.

For more information, including exposure control plans and rDNA information, go to EH&S' [Biosafety](#) web page.

**REFERENCES**

FAQs on Incident Reporting - web page  
<https://oas.od.nih.gov/biotechnology/freq-on-incident-reporting/>



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 08/2019

<http://tiny.cc/usc-rDNA-biohz-rpt>



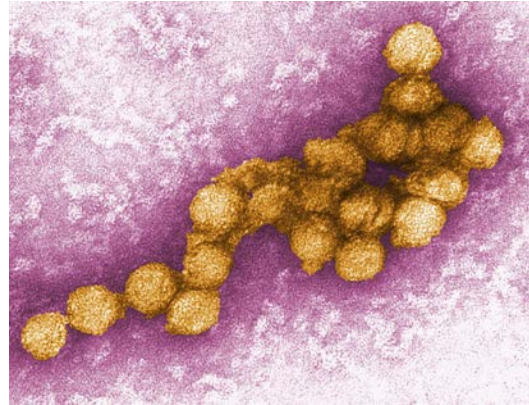
## 4.0 Biohazards

There are many types of biological materials used in research, each with its own hazards and risks. However, these hazards/risks may not be immediately apparent to the researcher. The following is a brief outline and description of the various biological materials that may be hazardous and the risks associated with them.

The USC IBC has a [policy](#) on the requirement for a Biohazardous Use Authorization (BUA). The following biomaterials are classified as biohazards and require a BUA:

- Recombinant DNA (rDNA) and synthetic nucleic acids
- Pathogenic microorganisms (see Figure 4.1)
- Human or non-human primate blood, tissue, and organs, or other potentially infectious material (OPIM)
- Human and non-human primate cell lines
- Toxins of biological origin
- Hazardous chemicals used in biomedical research
- Unknowns

**Figure 4.1. West Nile**



Source: CDC

## Recombinant and Synthetic Nucleic Acids

Research involving recombinant or synthetic nucleic acids (rDNA research) at USC is subject to guidelines set forth by the National Institutes of Health (NIH) in the *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules* (see Figure 4.2). Although titled “guidelines”, the rules set forth are required for any entity that accepts federal grant money and are thus *de facto* regulations.

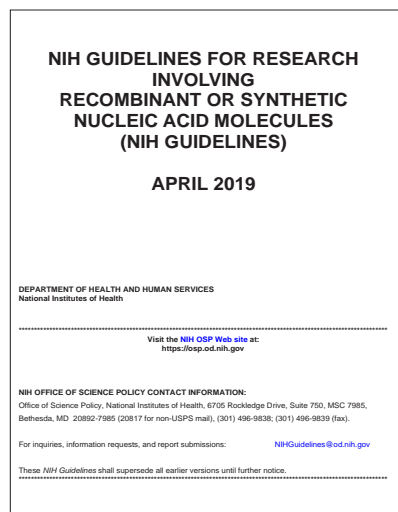
Definitions according to the NIH Guidelines regarding what is covered are the following:

- Recombinant nucleic acids – molecules that are constructed by joining nucleic acid molecules and can replicate in a living cell;
- Synthetic nucleic acids – nucleic acid molecules that are chemically or by other means synthesized or amplified, including those that are chemically or otherwise modified but can base pair with naturally occurring nucleic acid molecules; or
- Molecules that result from the replication of those described above.

Recombinant agents are considered biohazards. The risks are dependent upon the components of each recombinant construct.

The use of recombinant materials usually involves three major components: the vector, the gene(s) or other nucleic acid of interest, and the host, each with inherent risks. The combination of these three components can result in an agent that is more (or less) hazardous than the starting materials.

**Figure 4.2. NIH Guidelines for rDNA**



Source: NIH

Cloning **vectors** are used in rDNA technology as a vehicle for inserting a gene or other nucleic acid sequence into the host cell. There are many types of cloning vectors, but the most commonly used are plasmid vectors (e.g., pBR322, pUC19) and viral vectors (e.g., adenoviral vector, lentivirus vector).

**Genes** encode protein sequences to be transferred. Genes that may increase the risks associated with the final construct include those genes encoding toxins, oncogenic proteins, and physiological modifiers.

**Hosts** generally include bacteria, human or animal cell lines, and yeast cells.

## Pathogenic Microorganisms

Pathogenic microorganisms are biohazards that include: bacteria, viruses, fungi, parasites, and [prions](#). Each type of microorganism, and each genus, species, and perhaps subtype will have its own hazards and risk factors to consider when preparing to perform research. The Public Health Agency of Canada provides access to [Pathogen Data Sheets](#) for a number of microorganisms that affect human health.

The **virulence** of a pathogenic microorganism is a description of how well it replicates in its host, modifies the host defenses, allows the spread of the disease within the host, and is toxic to the host. These factors determine whether infection occurs and how severe the disease symptoms are.

A host's resistance is based on multiple factors including age, stress, and physiologic attributes.

**Morbidity** describes relative incidents of disease. In other words, morbidity is the number of cases of disease in a given area.

**Mortality** refers to the number of deaths in a specific population. Mortality rates are typically expressed as a percentage of people who die from the disease in the overall infected population. For example, the mortality rate for Ebola virus can be as high as 50%.

Mortality rates are typically considered a conservative rate as many cases of a disease may go unreported. This can be especially true in third world countries.

### Bacteria

Bacteria are prokaryotic cells that lack a membrane-bound nucleus, have no membrane-bound organelles, and possess cell walls. Bacteria are ubiquitous, found thriving in the harshest environmental conditions as well as being a vital part of human health in the form of the natural flora of the human body.

In fact, there are more than  $10^{14}$  bacteria in the average human body, more than the number of cells that make up the human body. Although bacteria are a vital part of the environment, many can cause disease.

Examples of pathogenic bacteria are:

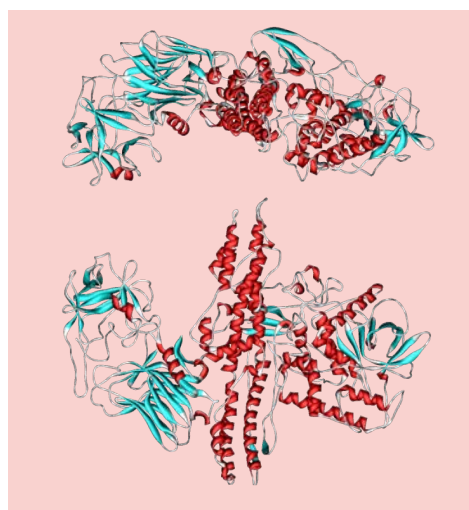
- *Staphylococcus aureus* which can infect skin and other parts of the body.
- *Bacillus anthracis* which produces anthrax spores.
- *Clostridium botulinum* which produces botulinum neurotoxins.
- *Salmonella typhimurium* which can cause gastrointestinal disorders.

**Bacterial spores.** Some bacteria can produce spores (endospores) during times of unfavorable conditions. The spore is a dormant cell that is resistant to harsh environments. When conditions are more favorable for the bacteria, the spore will differentiate to become the bacteria in a process called germination. Germination is not a form of reproduction, but differentiation into a different cell type that may then divide to form more bacteria.

Spores by nature are resistant to harsh conditions (e.g., heat, UV radiation, and desiccation) and, by virtue of this quality, resistant to typical decontamination processes. It is important to select the appropriate inactivation process for decontamination of spore-forming bacteria used in research.

**Bacterial Toxins.** There are two types of bacterial toxins, those that are secreted - exotoxins - and those that are retained by the bacterium - endotoxins. Endotoxins (lipopolysaccharides; LPS) are the structural components of Gram-negative cell walls of the bacterium and are released when the cell wall disintegrates. A familiar example of a toxin is botulinum neurotoxin (see Figure 4.3). It is secreted by *Clostridium botulinum*. This neurotoxin can cause a fatal disease called botulism, but in extremely small increments is a common tool in the aesthetics business, used to reduce wrinkles, or in neurology, as a tool for headache mediation. One commercial name for this product is Botox.

**Figure 4.3. Botulinum neurotoxin**



Source: Wikipedia

## Viruses

Viruses are metabolically inert, infectious agents that replicate only within the cells of living hosts such as bacteria, plants, and animals. These agents have a core composed of either RNA or DNA, a protein coat, and, in more complex virus types, have a surrounding envelope. The function of the viral particle or virion is to deliver its DNA or RNA package into the cell.

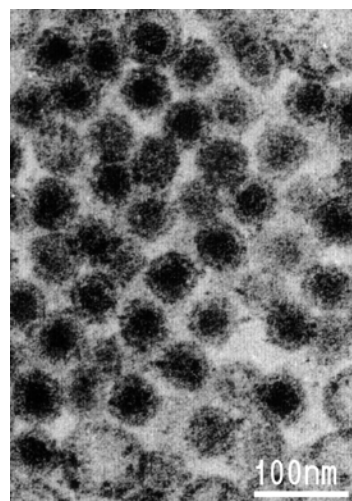
Once the delivery has taken place, a virus will supplant appropriate parts of the cellular machinery. The goal of this seizure is to produce viral proteins and ultimately viral particles to spread the infection.

Every virus has a typical or natural means of transmission to their hosts. Some are carried by a vector (living host) while others spread person-to-person directly or indirectly.

**Arboviruses** are viruses that infect a living arthropod carrier. The virus can spread after a person is bitten by an infected arthropod. Common carriers (vectors) are mosquitoes (insects) and ticks (arachnids).

Examples of arboviruses are West Nile virus, Dengue virus (see Figure 4.4), and Japanese Encephalitis.

**Figure 4.4. Dengue virus**



Source: The Japan Times

**Zoonotic viruses** are those that infect animal species in addition to humans (arboviruses are zoonotic viruses as well). Zoonotic viruses (e.g., Marburg virus; see Figure 4.5) typically infect a specific animal, but the disease can often be passed onto humans. Since humans are in constant contact with pet animals we keep in our homes, zoonotic diseases can be transmitted between pets and owners.

According to the CDC, it is estimated that six (6) out of ten (10) diseases (viral, bacterial, fungal, etc.) are spread from animals.

Transmission can occur in a variety of ways:

- Contact with infected bodily fluids (saliva, blood) or feces;
- Bites or scratches
- Eating unsafe or contaminated food products from animals such as unpasteurized milk and milk products.

Examples of zoonotic viruses are avian influenza virus, Junín virus, rabies virus, Ebola virus, and Marburg virus (see Figure 4.5).

Examples of zoonotic agents in rodents include viral agents such as Lymphocytic Choriomeningitis Virus (LCMV) or Hantavirus.

Department of Animal Resources veterinarians ensure that USC rodents are free from these and other zoonotic agents through strict animal health surveillance and screening of cell lines and other biologicals that may contain them. In addition, agents such as rabies virus are prevented by ensuring vaccination of susceptible carnivore animals.

**Figure 4.5. Marburg virus**



Source: Wikimedia Commons

Macaque nonhuman primates may harbor a zoonotic herpesvirus (commonly called B virus). Personnel handling these animals must be enrolled in a program for health surveillance and training in procedures for preventing exposure and for treatment in case of possible exposure.

**Pathogenicity** refers to the severity of the disease that follows infection. The mechanisms of pathogenesis in viruses include:

- Attachment and penetration of the virus at the site of entry
- Replication inside the host cell
- Spread of virus to target organs (disease sites)
- Spread to sites where shedding of the virus into the environment occurs

Some of the factors that affect pathogenic mechanisms are:

- Accessibility of virus to human or animal tissue
- Susceptibility of infected cells to virus multiplication
- Susceptibility of virus to host defenses.

## Fungi

Fungi are abundant worldwide and were classified into their own life kingdom separate from plants in 1969. Fungi are eukaryotic organisms and range from mushrooms to human pathogens. Fungi can cause different types of infections.

**Superficial** infections will occur on the outermost layer of the skin or hair shafts. No viable tissue is invaded and there is no cellular immune response by the host. These types of infections typically have mild to no symptoms. Infected individuals may not even be aware of the infection. Examples of superficial mycoses agents are:

- *Piedraia hortae* causes black piedra;
- *Hortaea werneckii* causes tinea nigra

**Cutaneous** infections occur with keratinized tissue such as skin, hair, and nails. These types of infections do not typically invade living tissue and will likely remain in the nonviable regions of the keratinized tissues. These infections are not debilitating or life-threatening, but they are often chronic in nature. Cutaneous mycoses will typically cause a cellular response in the host. Examples of cutaneous mycoses agents are:

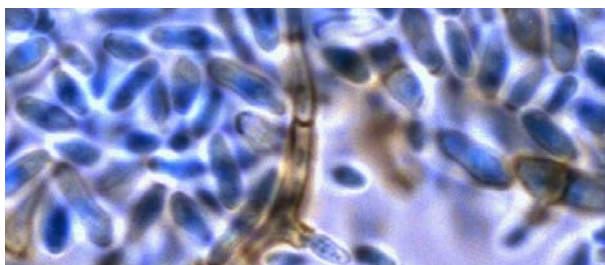
- Ringworm - caused by various species in the genera *Trichophyton* and *Microsporum*
- Athlete's foot - caused by *Tinea pedis*

**Subcutaneous** infections usually occur after a traumatic incident causing broken skin and providing a point of ingress to the fungal agent. Subcutaneous mycoses are rare conditions in humans. The causative fungal agents of these infections are soil fungi.

Examples of subcutaneous mycoses agents:

- *Loboa loboi*
- *Rhinosporidium seeberi* (see Figure 4.6)

**Figure 4.6. *Rhinosporidium seeberi***



Source: Alchetron

**Systemic** infections typically occur through the inhalation of fungal spores also known as conidia. The initial infection will likely take place in the lungs and can progress to other tissue from there. These are fungal agents that are dimorphic having two distinct morphologies, one of which is capable of survival within a human host. Systemic mycoses agents can pose serious health hazards in laboratories if inhaled. Some examples are:

- *Blastomyces dermatitidis*
- *Coccidioides immitis*
- *Histoplasma capsulatum*

**Opportunistic** infections occur with ubiquitous fungi that would not typically cause illness in healthy human adults. These fungi are typically part of the human body's normal flora but can cause disease in people with immunodeficiencies or who are immunocompromised. Examples of opportunistic mycoses include:

- Candidiasis - typically caused by *Candida albicans*
- Cryptococcosis - caused by *Cryptococcus neoformans*
- Aspergillosis - often caused by *Aspergillus fumigatus*

## Parasites

Parasites are organisms that live in or on another organism known as the host. The parasite acquires its nutrients at the expense of the host. Parasites include:

- Protozoa (parasitic single cell organisms: *Plasmodium spp.*, malaria. See Figure 4.7)
- Arthropods (parasitic insects and arachnids e.g., ticks)
- Helminthes (parasitic worms e.g., tapeworms)

Parasites do not typically kill their hosts since that is detrimental to their own survival, but infections do reduce the general health of the host. Parasites can be classified based on their life cycles or their interactions with a host. A simple classification can be where the parasite lives either on the surface or interior of its host.

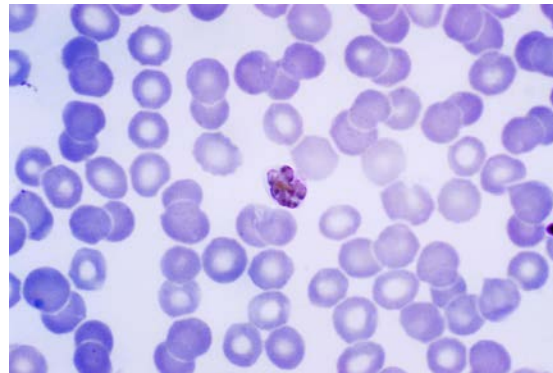
**Ectoparasites** live on the surface of the host. Arthropods such as ticks and mites are ectoparasites.

**Endoparasites** live within their host for some portion during their life cycle. Typical endoparasites are helminthes and protozoa.

**Protozoa** are unicellular eukaryotic organisms. They are typically classified by their means of motility, or their location in their host. Most protozoa infect blood or the intestines of their hosts. A few examples are provided below.

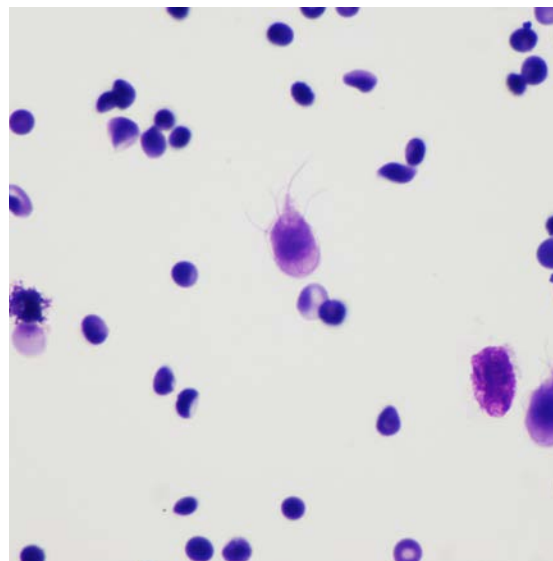
**Intestinal protozoa.** Intestinal infections are caused by various protozoa including *Entamoeba histolytica*, *Giardia intestinalis* (lamblia) (see Figure 4.8), *Cystoisospora* (*Isospora*) *belli*, *Cyclospora cayetanensis*, or *Cryptosporidium* (most commonly spread by municipal water and recreational water).

**Figure 4.7. *Plasmodium malariae***



Source: Wikipedia

**Figure 4.8. *Giardia intestinalis***



Source: Wikimedia Commons

Note that nonpathogenic commensal protozoa can be present at the same time.

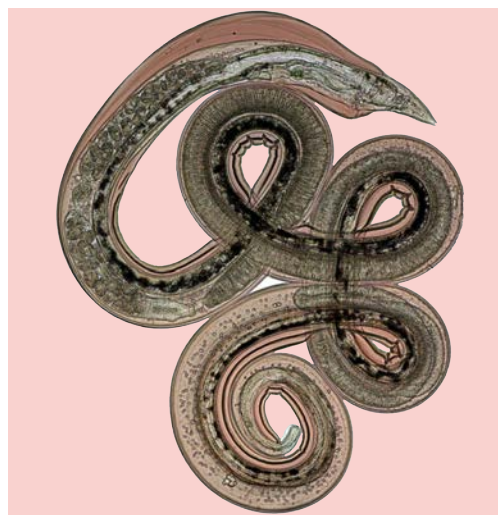
**Systemic protozoa** or bloodborne protozoa include bloodborne parasites that are spread via insect vectors such as mosquitoes. Malaria is the most well-known bloodborne parasitic disease being caused by various species of the genus *Plasmodium*.

**Helminthes** (e.g., *Nippostrongylus brasiliensis*; see Figure 4.9) are parasites that can typically be seen with the naked eye in their adult form. They are often referred to as “intestinal worms” although not all will reside in the intestines. They all share the common characteristic of being worm-shaped, but they are all genetically quite diverse from one another. Some common groups are listed below.

**Tapeworms** are parasites that live within the intestinal track of their hosts. Infection generally occurs through the consumption of undercooked meats or foods prepared in unsanitary conditions. These worms will grow within the intestinal tract but usually, do not cause symptoms in the host.

**Roundworms** also reside in the intestines of their host. Symptoms vary from none to significant usually due to the amount of worms present. Infection will generally occur in areas with poor sanitation or in people with poor personal hygiene. Hookworm (*Ancylostoma duodenale* and *Necator americanus*) and pinworm (*Enterobius vermicularis*) infections are examples of common human roundworm infestations.

**Figure 4.9.** *Nippostrongylus brasiliensis*



Source: Worldwide Universities Network

**Flukes** (or Flatworms) have two hosts during their lifecycle: one that is typically a snail with the other a vertebrate. *Schistosoma spp.* (see Figure 4.10) represent a group of flukes that pose significant health concerns causing such diseases as snail fever and schistosomiasis.

Transmission typically occurs through water in which the swimming larvae bore through human or animal skin.

## Prions

Transmissible spongiform encephalopathies (TSE) or prions are infectious agents composed entirely of protein material called PrP (proteinaceous infectious particles) that cause neurodegenerative diseases. Prions (see Figure 4.11) are difficult to inactivate due to their structure.

The biochemical feature of prion diseases is the conversion of a normal prion protein (PrP) to an abnormal, misfolded, pathogenic isoform called PrP<sup>Sc</sup> (Sc is for “scrapie” the prototypical prion disease).

The conformational changes associated with PrP<sup>Sc</sup> are an increase of  $\beta$ -pleated sheets and a reduction of  $\alpha$ -helixes. These  $\beta$ -pleated sheets allow for the accumulation of the PrP<sup>Sc</sup> in the central nervous system resulting in spongiform (microscopic sponge-like appearance of the brain tissue) and ultimate neurodegeneration.

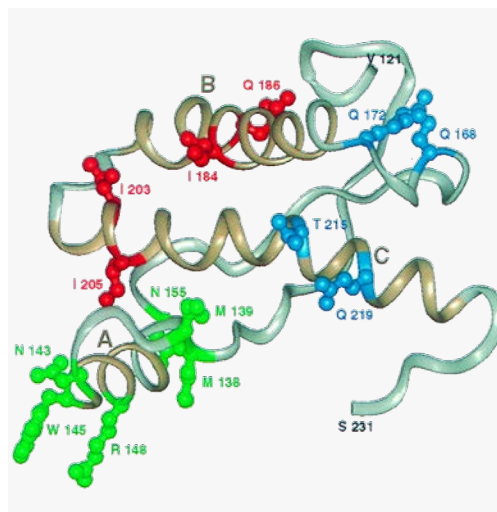
Prion diseases affect both humans and animals such as sheep and cattle. There are no therapies or vaccines for any of the prion diseases. The incubation period can be from a few months to a few decades. Once the disease manifests, it is typically fatal within one year.

**Figure 4.10. *Schistosoma mansoni***



Source: Wikipedia

**Figure 4.11. Prion Protein**



Source: Proceedings of the National Academy of Sciences

The prion replication method does not include genetic material (DNA or RNA) associated with the prion proteins. The misfolded prion proteins self-propagate causing protein accumulation.

Familial Creutzfeldt-Jakob disease (CJD) accounts for 15% of diagnosed prion disease in humans and is passed on in an autosomal dominant manner according to the CDC.

The following are important characteristics when reviewing the risks associated with a microorganism:

- Fatality rate
- Host range
- Infectious dose
- Mode of transmission
- Incubation period
- Viability
- Communicability
- Dissemination
- Available medical interventions

Consider each characteristic carefully and decide how to protect yourself from the risks particular to the agent.

## Toxins of Biological Origin

Toxins of biological origin are toxic substances produced by microorganisms, animals, and plants that have the capability of causing harmful effects when inhaled, ingested, injected, or absorbed by humans or other mammals. The LD<sub>50</sub> is an abbreviation for "Lethal Dose, 50%" or median lethal dose. It is the amount of the substance required (usually per body weight) to kill 50% of the test population.

The term toxin is sometimes used improperly to refer to any toxic material. Toxicant refers to any toxic substance, whereas, toxin is a toxicant of biological origin. Some toxins are select agents and must be used in accordance with CDC guidelines.

There are exemptions from select agent registration given for toxins if the amount held by any one principal investigator is under the permissible amount. Table 4.1 indicates the permissible amounts as of December 2018.

**Table 4.1. Permissible Toxin Amounts of Select Agents**

HHS Toxins [§73.3(d)(7)]	Amount (mg)
Abrin	1000
Botulinum neurotoxins	1
Short, paralytic alpha conotoxins	100
Diacetoxyscirpenol (DAS)	10,000
Ricin	1000
Saxitoxin	500
Staphylococcal Enterotoxins (Subtypes A, B, C, D, and E)	100
T-2 toxin	10,000
Tetrodotoxin	500

<https://www.selectagents.gov/PermissibleToxinAmounts.html>

## Hazardous Chemicals of Biomedical Research

- Carcinogens, mutagens, and teratogens
  - The [California Proposition 65 List](#) reports chemicals that are "known to cause cancer or birth defects or other reproductive harm."
- Chemotherapy agents - cytotoxic by design

The USC IBC requires registration of the above chemicals to ensure that all researchers are aware of the hazards associated with them. For more information regarding the use of hazardous chemicals, refer to the **Chemical Hygiene Plan**.

## Unknowns

"Unknowns" is a catch-all category assigned to biological material whose hazard determination is not yet established. The Universal Precautions concept is invoked when handling unknown biological materials (see below) i.e., treat the biomaterials as though they are potentially infectious.

- Microorganisms collected in the environment.
- Animal tissues, carcasses, or fluids collected in the environment
- Pathological specimens
- Any human or non-human primate materials

## Select Agents

USC is registered with the Federal Select Agent Program (FSAP) that regulates the possession, use, and transfer of biohazardous select agents and toxins. This program is highly regulated by both the CDC and the USDA. Each Principal Investigator and each agent must be separately registered with the select agent program.

The EH&S Biosafety Program can assist you if you are thinking of working with select agents. More information about this program can be found online at <https://www.selectagents.gov/>.

See [Appendix A](#) for a complete list of select agents and toxins that is current for 2018.

## Dual Use Agents of Concern (DURC)

Dual Use Research of Concern (DURC) is defined in the United States Government (USG) policy statement as "life sciences research that, based in current understanding, can be reasonably anticipated to provide knowledge, information, products, or

technologies that could be directly misapplied to pose a significant threat with broad potential consequences to public health and safety, agricultural crops and other plant, animals, the environment, materiel, or national security."

There are fifteen agents and toxins and seven activities that constitute DURC research. The agents and toxins and the activities may be found on the EH&S website <https://ehs.usc.edu/files/ehs-durc-10-2015.pdf>.

Once a PI identifies as using one of the fifteen agents, the EH&S Biosafety Office in conjunction with the USC IBC will assist with the determination regarding whether or not the activities meet the definition given in the policy.

Ultimately, DURC research must be reported to, and vetted by, the funding agency. This may require extra time so if a researcher believes that the project will involve DURC, registering the BUA with the USC IBC well in advance is advisable.

The USC Office of EH&S Biosafety Program has produced the [USC DURC Manual](#) in compliance with the United States Government Policy for Institutional Oversight of Life Sciences Dual Use Research of Concern.

See <http://www.phe.gov/s3/dualuse> for more information about this program.

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## 5.0 Principles of Biosafety

The overarching purpose of the practice of biosafety is to find methods to contain biological hazards. Containment of biohazards will reduce or eliminate the risks to humans, animals and plants, and the environment. Understanding the fundamental principles of biosafety will lead to a safer and more comfortable work environment for all USC employees.

### Risk Assessment

One of the basic tenets of biosafety is to perform a risk assessment prior to beginning work on a new project involving biological agents. Performing a risk assessment will provide a good idea about the hazards involved in a project, what risks are associated with each hazard, and how the risks can be addressed to limit injury and illness.

A risk assessment involves the following steps:

- Identification of hazards;
- Determination of risks associated with each hazard;
- Determination of both probability and consequences of each risk; and
- Finding a means to eliminate or reduce the identified risks.

Hazard and risk are sometimes used interchangeably but they do not mean the same thing. A hazard is something that causes harm or an adverse effect. A risk is the probability of the hazard causing harm and the severity of the harm caused. Risk assessment (See Table 5.1) helps determine what biosafety containment level is necessary to work with and handle the biological agents.

**Table 5.1. Risk Assessment Matrix**

Probability		Consequence				
		Injuries/ Ailments - No medical treatment	Minor Injuries/ First Aid	Serious injury/ Hospitalization	Life threatening/ Multiple serious injuries	Death/ Multiple life-threatening injuries
		Insignificant	Minor	Moderate	Major	Catastrophic
		1	2	3	4	5
Almost Certain	5	M	H	H	E	E
Likely	4	M	M	H	H	E
Possible	3	L	M	M	H	E
Unlikely	2	L	M	M	H	H
Rare	1	L	L	M	M	H

E - Extreme Risk; Detailed action plan required to manage risk H - High Risk; Immediate attention needed

M - Medium Risk; Specify responsibilities L - Low Risk; Manage by routine procedures

## Risk Groups and Biosafety Levels

Microorganisms are classified by risk groups. The classification of microorganism takes into account the characteristics of the agent such as:

- Infectious dose
- Pathogenicity
- Mode of transmission
- Host range
- Preventive therapies or current treatments

There are four risk groups: Risk Group 1 (RG1), the lowest risk group through Risk Group 4 (RG4), the highest risk group. The risk groups of different microorganisms are classified and defined by the NIH Guidelines, the WHO Laboratory Biosafety Manual, and the *Biosafety in Microbiological and Biomedical Laboratories 5th Edition*. Table 5.2 provides an overview of the four risk groups.

Biosafety levels (BSLs) are the handling and containment levels for biological materials and are consistent with Risk Groups, in most cases. However, the way a microorganism is used, its location in the environment, and other factors are considered when determining the biosafety level. Risk groups are: (a) determined by microbiologists and biomedical researchers based on aggregate data, (b) universally agreed upon, and (c) independent of how the agent is used; Biosafety Level is determined by the Institutional Biosafety Committee (IBC) based on the agent used and how it will be manipulated.

Table 5.3 details handling practices and containment for the four Biosafety Levels. For more complete descriptions of the biosafety levels used with biological agents, see CDC/NIH publication [Biosafety in Microbiological and Biomedical Laboratories 5th Edition](#). For a complete description of the biosafety levels used with recombinant microorganisms, large scale (greater than 10 L) research, plants, or large animals, see The [NIH Guidelines for Recombinant and Synthetic Nucleic Acid Molecules, Appendices G, K, P, and N](#).

**Table 5.2. Classification of Infectious Microorganisms by Risk Group**

<b>Risk Group Classification</b>	<b>NIH Guidelines<sup>1</sup></b>	<b>WHO<sup>2</sup></b>
Risk Group 1	Agents not associated with disease in healthy adult humans	(No or low individual and community risk) A microorganism unlikely to cause human or animal disease.
Risk Group 2	Agents associated with human disease that is rarely serious and for which preventive or therapeutic interventions are often available.	(Moderate individual risk; low community risk) A pathogen that can cause human or animal disease but is unlikely to be a serious hazard to laboratory workers, the community, livestock or the environment. Laboratory exposures may cause serious infection, but effective treatment and preventive measures are available and the risk of spread of infection is limited.
Risk Group 3	Agents associated with serious or lethal human disease for which preventive or therapeutic interventions may be available.	(High individual risk; low community risk) A pathogen that usually causes serious human or animal disease but does not ordinarily spread from one infected individual to another. Effective treatment and preventive measures are available.
Risk Group 4	Agents likely to cause serious or lethal human disease for which preventive or therapeutic interventions are not usually available.	(High individual and community risk) A pathogen that usually causes serious human or animal disease and can be readily transmitted from one individual to another, directly or indirectly. Effective treatment and preventive measures are not usually available.

1 - NIH Guidelines for Research involving Recombinant DNA Molecules 2002; 2 - World Health Organization Laboratory Biosafety Manual 3rd Edition 2004; Biosafety in Microbiological and Biomedical Laboratories 5th Edition 2009

**Table 5.3. Biosafety Levels**

<b>BSL</b>	<b>Practices</b>	<b>Primary Barriers &amp; Safety Equipment</b>	<b>Facilities (Secondary Barriers) &amp; Special Equipment</b>
<b>BSL 1</b>	Standard microbiological practices	<ul style="list-style-type: none"> <li>No primary barriers needed</li> <li>PPE may include lab coats, gloves, eye and face protection as needed</li> </ul>	Lab bench and sink required
<b>BSL 2</b>	BSL1 practices plus: <ul style="list-style-type: none"> <li>Limited access</li> <li>Biohazard warning signs</li> <li>Sharps precautions</li> <li>Biosafety manual with waste decontamination or medical surveillance policies</li> </ul>	<ul style="list-style-type: none"> <li>BSCs or other containment equipment to manipulate infectious agents that may have splash or aerosolization hazards</li> <li>PPE may include lab coats, gloves, eye and face protection as needed</li> </ul>	BSL1 requirements plus Autoclave available
<b>BSL 3</b>	BSL2 practices plus: <ul style="list-style-type: none"> <li>Controlled access</li> <li>Decontamination of all waste</li> <li>Decontamination of all lab clothing before laundering</li> </ul>	<ul style="list-style-type: none"> <li>BSCs or other containment equipment for all open manipulations of infectious agents</li> <li>PPE may include lab coats, gloves, eye and face protection, and respiratory protection as needed</li> </ul>	BSL2 requirements plus: <ul style="list-style-type: none"> <li>Physical separation from access corridors</li> <li>Self-closing, double door access</li> <li>Exhausted air not recirculated</li> <li>Negative airflow into laboratory</li> <li>Entry through airlock or anteroom</li> <li>Hand washing sink near laboratory exit</li> </ul>
<b>BSL 4</b>	BSL3 practices plus: <ul style="list-style-type: none"> <li>Clothing change before entering lab</li> <li>Shower required on exit</li> <li>All material must be decontaminated on exit from facility</li> </ul>	<ul style="list-style-type: none"> <li>All procedures conducted in ducted BSCs in combination with full body, air supplied, positive pressure air suit.</li> </ul>	BSL3 requirements plus: <ul style="list-style-type: none"> <li>Separate building or isolated zone</li> <li>Dedicated supply and exhaust, vacuum, and decontamination systems</li> <li>Other requirements as stated in BMBL</li> </ul>

Reference: Biosafety in Microbiological and Biomedical Laboratories 5th Edition



## 6.0 Animals and Animal Biosafety Levels

There are a number of hazards associated with laboratory animals. These include allergens, bites and scratches, zoonoses, as well as the chemicals and infectious microorganisms with which animals may be treated in research. Some studies have shown that up to 30% of those who work with laboratory animals will develop allergies within the first three years of work (see Figure 6.1 Animal Allergies Fact Sheet). Allergies can lead to more serious diseases such as asthma, which can be life-threatening. There are other hazards associated with the animal facility itself, such as cleaning compounds, and the physical hazards associated with autoclaves, cage washers, and animal housing racks. The Office of Environmental Health and Safety's Biosafety Program developed the Occupational Medicine Animal Exposure Program, which is outlined in [Section 11 Occupational Medicine for Biological Research](#).

Figure 6.1. Animal Allergies Fact Sheet

### FactSheet Animal Allergies

**M**ost employees working in animal facilities find it similar to any other work environment. However, some employees may experience allergy symptoms that can be directly related to their work with or proximity to animals. Allergies can develop and the symptoms expressed even when there is no direct contact with animals.

**WHY DO ANIMALS CAUSE ALLERGIES?**  
Proteins in animal dander and excrement (urine and feces) may cause allergic reactions. Proteins that cause allergies are called "allergens" and may exist as airborne particles in animal facilities.

**CAN ANYONE BECOME ALLERGIC TO LABORATORY ANIMALS?**  
Individuals that have existing allergies, a family history or susceptibility to allergens, or suppressed immune systems may be at a higher risk for developing allergies. Others may develop allergies over time that increase in intensity as exposure increases; some may never experience allergies.

**WHAT ARE THE SYMPTOMS OF ANIMAL ALLERGIES?**  
Allergy symptoms consist of the following: itchy eyes, nasal stuffiness, sneezing, coughing, wheezing, difficulty breathing, a tight chest, and rashes on the skin. These symptoms can range from mild to severe.

**HOW DO I PROTECT MYSELF FROM ANIMAL ALLERGIES?**  
Three levels of control are employed to prevent workers from contact with animal allergens: engineering controls, workplace practices or administrative controls, and personal protective equipment (PPE).

**Engineering controls.** For USC animal facilities, these include: biosafety cabinets, animal transfer stations, cage dumping stations, and individually ventilated caging systems. This HEPA-filtered containment equipment is maintained and calibrated to industry standards.

Air quality monitoring of high risk areas is provided by EH&S. Personal and area monitoring may be performed to determine the effectiveness of controls.

**WHAT I NEED TO DO**

- Know the signs and how to report in case of a suspected allergic reaction.
- Use engineering and administrative controls as well as appropriate PPE to reduce exposure to animal allergies.
- Complete an [ADA Form](#) (see [Administrative controls](#) below).
- Wash hands after removal of protective gloves.

**Administrative controls.** These include the [Animal Exposure Program](#) and [Occupational Medicine for Biomedical Research](#). An [Animal Exposure Risk Assessment form](#) (ARA) that includes animal hazards, medical history, and factors that may influence allergy development is completed by the employee. An Occupational Medicine physician then reviews the completed ARA form. Based on the assessment, the following may be recommended: use of an N95 respirator, a follow-up evaluation with a physician, or both (refer to the [ADA Fact Sheet](#) for details).

**PPE.** The Department of Animal Resources (DAR) has a minimum PPE requirement for entering animal facilities that includes:

- Pants or other leg covering
- Closed-toe/closed heel shoes worn with socks
- Disposable, closed front gown
- Gloves
- Shoe covers
- Face mask
- Hair bonnet

Clothing that does not meet or exceed DAR requirements is not allowed in animal facilities.

**REFERENCES**

- [Occupational Health and Safety in the Care and Use of Research Animals \(OLC-2021\)](#)
- [Guide for the Care and Use of Laboratory Animals \(NIH, 2011\)](#)
- [NIH/OD: Involving Animals in Animal Studies](#)

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<http://tiny.cc/usc-ehs-AA-fs>

Many different animal species can be used for research. Some common examples of animals used in research include mice, rats, zebrafish, birds, guinea pigs, large animals such as rabbits, swine, and macaques (also referred to as non-human primates), and transgenic animals.

Transgenic animals are defined as animals that have a foreign gene inserted into their genome or a gene removed (knock-out). This is primarily done with mice. Transgenic mice may fall under the *The NIH Guidelines for Recombinant and Synthetic Nucleic Acid Molecules*. Answers to Frequently Asked Questions (FAQs) on transgenic animals is available at the [FAQs for Research Subject to the NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules \(NIH Guidelines\): Genetically Modified \(Transgenic\) Animals and the Use of Recombinant or Synthetic Nucleic Acid Molecules in Animals](#) web page.

## Zoonoses

Zoonotic agents are carried via living animal vectors. Zoonotic agents typically infect a specific animal, but the disease can often be passed onto humans. Humans are in constant contact with pet animals we keep in our homes so zoonotic diseases are quite common. According to the CDC, it is estimated that six out of ten diseases (viral, bacterial, fungal, etc.) are spread from animals.

Transmission can occur in a variety of ways:

- Contact with infected bodily fluids (saliva, blood) or feces;
- Bites or scratches
- Eating unsafe or contaminated food products from animals such as unpasteurized milk.

Examples of zoonotic agents are avian influenza virus, Junín virus, rabies virus, *Leptospira* spp., *Francisella tularensis*, *Chlamydia psittaci*, and *Trichinella* spp.

## Animal Cell Lines

Animal cell lines can also be used in biomedical research. They are generally considered risk group 1 agents. However, non-human primate (NHP) cells, both primary (isolated directly from NHP tissue using enzymatic or mechanical methods) and established (acquired ability to proliferate indefinitely), and any tissue from NHPs, are considered potentially infectious materials and should be handled as such.

## Animal Biosafety Levels

Animal Biosafety Levels (ABSL) are similar to Biosafety Levels but are specifically used for areas of containment when conducting research in animals. The BMBL stipulates what is required in order to establish the four levels of animal biosafety. ABSL1 is the lowest level of containment and ABSL4 is the highest level of containment. Table 6.1 briefly describes the ABSL requirements.

**Table 6.1. Animal Biosafety Levels**

BSL	Practices	Primary Barriers & Safety Equipment	Facilities (Secondary Barriers) & Special Equipment
ABSL 1	Standard animal care and management practices, including appropriate medical surveillance programs	As required for normal care of each species <ul style="list-style-type: none"><li>• PPE: laboratory coats and gloves; eye, face protection, as needed</li></ul>	Standard animal facility: <ul style="list-style-type: none"><li>• No recirculation of exhaust air</li><li>• Directional air flow recommended</li><li>• Hand washing sink is available</li></ul>

BSL	Practices	Primary Barriers & Safety Equipment	Facilities (Secondary Barriers) & Special Equipment
ABSL 2	ABSL-1 practice plus: <ul style="list-style-type: none"> <li>Limited access</li> <li>Biohazard warning signs</li> <li>"Sharps" precautions</li> <li>Biosafety manual</li> <li>Decontamination of all infectious wastes and animal cages prior to washing</li> </ul>	ABSL-1 equipment plus: <ul style="list-style-type: none"> <li>Containment equipment appropriate for animal</li> <li>PPE: Laboratory coats, gloves, face, eye and respiratory protection, as needed</li> </ul>	ABSL-1 facility requirements plus: <ul style="list-style-type: none"> <li>Autoclave available</li> <li>Hand washing sink available</li> <li>Mechanical cage washer recommended</li> <li>Negative airflow into animal and procedure rooms recommended</li> </ul>
ABSL 3	ABSL-2 practice plus: Controlled access <ul style="list-style-type: none"> <li>Decontamination of clothing before laundering</li> <li>Cages decontaminated before bedding is removed</li> <li>Disinfectant foot bath as needed</li> </ul>	ABSL-2 equipment plus: <ul style="list-style-type: none"> <li>Containment equipment for housing animals and cage dumping activities</li> <li>Class I, II or III BSCs available for manipulative procedures (inoculation, necropsy) that may create infectious aerosols</li> <li>PPE: Appropriate respiratory protection</li> </ul>	ABSL-2 facility requirements plus: <ul style="list-style-type: none"> <li>Physical separation from access corridors</li> <li>Self-closing, double-door access</li> <li>Sealed penetrations</li> <li>Sealed windows</li> <li>Autoclave available in facility</li> <li>Entry through ante-room or airlock</li> <li>Negative airflow into animal and procedure rooms</li> <li>Hand washing sink near exit of animal or procedure room</li> </ul>
ABSL 4	ABSL-3 practices plus: <ul style="list-style-type: none"> <li>Entrance through change room where personal clothing is removed and laboratory clothing is put on; shower on exiting</li> <li>All wastes are decontaminated before removal from the facility</li> </ul>	ABSL-3 equipment plus: <ul style="list-style-type: none"> <li>Maximum containment equipment (i.e., Class III BSC or partial containment equipment in combination with full body, air-supplied positive-pressure suit) used for all procedures and activities</li> </ul>	ABSL-3 facility requirements plus: <ul style="list-style-type: none"> <li>Separate building or isolated zone</li> <li>Dedicated supply and exhaust, vacuum, and decontamination systems</li> <li>Other requirements outlined in the text</li> </ul>

For more complete descriptions of the animal biosafety levels used with biological agents in animals, see CDC/NIH publication [Biosafety in Microbiological and Biomedical Laboratories 5th Edition](#).

## Department of Animal Resources

Laboratory personnel must be trained properly when handling animals in research. This includes species-specific training on handling and restraint, surgery and blood draw techniques, the basics for animal maintenance and experimental work, and any additional training requested.

The Department of Animal Resources, or DAR, is responsible for training in these areas. More information can be found at <https://research.usc.edu/animal-resources/>. The Biosafety Staff work very closely with the DAR staff to ensure safe procedures are followed.

### DAR/Biosafety Meetings

Research groups whose projects involve biohazards in animals must attend a DAR/Biosafety meeting prior to initiating any work. The meetings are mandated by the IBC, noted in the BUA approval, and scheduled through the Biosafety Program. USC veterinarians and Biosafety Team members will meet with research groups to review all biosafety and animal welfare aspects of the projects. Contact [biosafety@usc.edu](mailto:biosafety@usc.edu) to schedule a meeting.

## Institutional Animal Care and Use Committee

Anyone who wants to conduct research with animals must get approval from the Institutional Animal Care and Use Committee (IACUC). The application can be completed via the iStar system - <https://istar.usc.edu/iStar/PublicCustomLayouts/SSO/Selection>. This is the same online application that is used for IBC registration. More information on IACUC can be found at <https://iacuc.usc.edu/>.

The Biosafety Staff work very closely with the IACUC to facilitate the approval process in a timely manner.



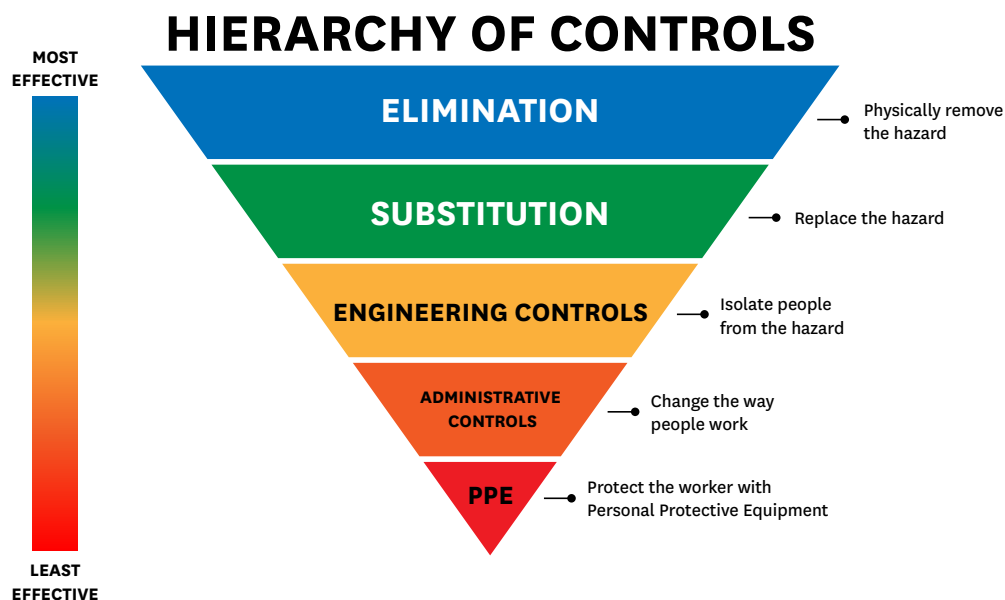
## 7.0 Biosafety Practices

It is very important to eliminate risks associated with the handling of biological materials wherever possible. This is accomplished through the implementation of Hazard Control Measures.

### Hazard Control Measures

Hazard control measures are used to reduce the risks associated with biomedical research. Control measures can be viewed in a hierarchal fashion (see Figure 7.1).

*Figure 7.1 Hierarchy of Controls*



The hierarchy of controls ranks measures from most effective to least effective. Elimination, substitution, engineering, administrative, and personal protective equipment are the commonly accepted control measures.

### Elimination

Elimination is the removal of the hazard from use. The use of synthetic blood instead of potentially infectious human blood in a teaching laboratory results in the elimination of a hazard.

## Substitution

Substitution as a control measure is the use of a more benign form of the hazard. For example, using a nonpathogenic strain instead of a wild type pathogen reduces the risk but does not completely eliminate it.

## Engineering Controls

OSHA defines engineering controls as “methods that are built into the design of a plant, equipment, or process to minimize the hazard.” Engineering controls are the first line of defense against hazards. They must be considered first when determining how to safely conduct a research project.

Ventilation is used as an effective engineering control in laboratory design. Air flows into a research laboratory and exits via an exhaust duct creating negative air flow relative to the outside; no air is recirculated back into the lab. Additionally, the laboratory experiences at least eight to ten room air changes per hour to remove airborne hazards.

An engineering control used in biomedical research is the biosafety cabinet (see Figures 8.1 and 8.2). BSCs are one-person workstations that use filtered air to protect the user from exposure and the biomaterial from contamination. See [Section 8 Laboratory Equipment](#) for more details on biosafety cabinets.

Other examples of engineering controls are: HEPA filters on vacuum lines, safety-engineered needles (see Figure 12.4), centrifuge safety cups and O-rings on rotor covers; automatic pipettes and sharps containers.

## Administrative Controls

Written safety policies, standard operating procedures (SOPs), safety training, and medical surveillance are all primary examples of administrative controls:

- SOPs are comprised of procedural steps, safe work practices, safety information (e.g., Safety Data Sheets - SDS), and emergency response. They are prepared by the PI, Lab Manager, or designee.
- Safety Training (e.g., BBP, SOP, general safety, and annual refresher) is necessary to prepare researchers prior to working in the lab. All training must be documented by the provider and records secured for at least three years. For more information on research safety training, visit: [Research Safety Training](#).
- Employees exposed to bloodborne pathogens either directly or indirectly are given access to medical surveillance in the form of the [Hepatitis B Immunization Program](#). Employees working with animals participate in the [Animal Exposure Program](#).

## Personal Protective Equipment

The [USC Personal Protective Equipment Standards](#) (see Figure 7.2) states that anyone entering an area that contains hazardous materials including chemicals, biological agents, and radiological materials or hazardous equipment must:

- At a minimum, wear full-length pants (or clothing that otherwise fully covers the legs and ankles) and closed toe/heel shoes;
- As determined by hazard assessment, wear hazard appropriate laboratory coats or equivalent garments, ANSI-approved protective eyewear, and other appropriate PPE (such as gloves and eye protection) when working with hazardous materials.
- Appropriate PPE is determined by completion of the [Lab Hazard Assessment Tool \(LHAT\)](#). For example, if the researcher indicates on the LHAT that (s)he works with human or non-human primate blood, body fluids, tissues, cells, or other potentially infectious materials (OPIM), then a fluid resistant (aka barrier) lab coat is needed; the fluid resistant lab coat is 100% polyester. Eye and face protection are required when splash hazards are present. The LHAT is required to be completed annually or when new hazards are introduced into the laboratory.

For information on selection of appropriate eye protection, consult the [Eye Protection Fact Sheet](#) (see Figure 7.3). For information on lab coat selection, consult the [Personal Protective Equipment \(PPE\): Lab Coat Selection Fact Sheet](#) (see Figure 7.4). Contact [biosafety@usc.edu](mailto:biosafety@usc.edu) for information on selecting the proper gloves, foot protection, eyewear, and face shields.

Figure 7.2 USC Personal Protective Equipment Standards

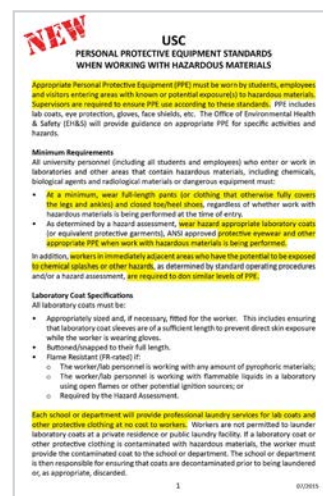
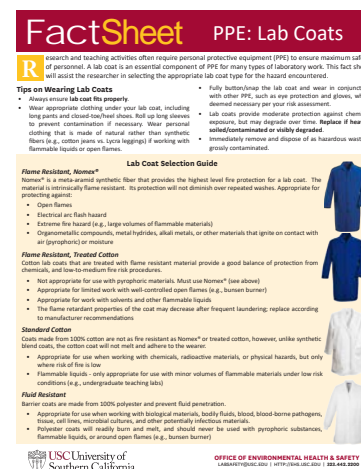


Figure 7.3. Eye Protection Fact Sheet



Figure 7.4. Lab coats Fact Sheet



## Standard Microbiological Practices

Each laboratory in which biological materials are handled must develop and train on procedures, practices, and precautions that are suitable for that laboratory, whether the agents used are risk group 1, 2, or 3. However, the CDC/NIH publication *Biosafety in Microbiological and Biomedical Laboratories 5th Edition* outlines standard microbiological practices that are highly recommended for all researchers to observe in addition to agent-specific procedures.

Standard Microbiological Practices are accepted as the initial standard of practice and are built upon depending upon the specific materials used.

1. The laboratory supervisor must enforce the institutional policies that control access to the laboratory.
2. Persons must wash their hands after working with potentially hazardous materials and before leaving the laboratory.
3. Eating, drinking, smoking, handling contact lenses, applying cosmetics, and storing food for human consumption must not be permitted in laboratory areas. Food must be stored outside the laboratory area in cabinets or refrigerators designated and used for this purpose.
4. Mouth pipetting is prohibited; mechanical pipetting devices must be used.
5. Policies for the safe handling of sharps, such as needles, scalpels, pipettes, and broken glassware must be developed and implemented. Whenever practical, laboratory supervisors should adopt improved engineering and work practice controls that reduce risk of sharps injuries. Precautions, including those listed below, must always be taken with sharp items.

These include:

- a. Careful management of needles and other sharps are of primary importance. Needles must not be bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated by hand before disposal.
  - b. Used disposable needles and syringes must be carefully placed in conveniently located puncture-resistant containers used for sharps disposal.
  - c. Non-disposable sharps must be placed in a hard walled container for transport to a processing area for decontamination, preferably by autoclaving.
  - d. Broken glassware must not be handled directly. Instead, it must be removed using a brush and dustpan, tongs, or forceps. Plasticware should be substituted for glassware whenever possible.
6. Perform all procedures to minimize the creation of splashes and/or aerosols.
  7. Decontaminate work surfaces after completion of work and after any spill or splash of potentially infectious material with appropriate disinfectant.
  8. Decontaminate all cultures, stocks, and other potentially infectious materials before disposal using an effective method.

Depending on where the decontamination will be performed, the following methods should be used prior to transport:

- a. Materials to be decontaminated outside of the immediate laboratory must be placed in a durable, leak proof container and secured for transport.
  - b. Materials to be removed from the facility for decontamination must be packed in accordance with applicable local, state, and federal regulations.
9. 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 (or other responsible personnel), telephone number, and required procedures for entering and exiting the laboratory. Agent information should be posted in accordance with the institutional policy.
  10. An effective integrated pest management program is required. This is provided by FMS Pest Control.
  11. The laboratory supervisor must ensure that laboratory personnel receive appropriate training regarding their duties, the necessary precautions to prevent exposures, and exposure evaluation procedures. Personnel must receive annual updates or additional training when procedural or policy changes occur. Personal health status may impact an individual's susceptibility to infection, ability to receive immunizations or prophylactic interventions. Therefore, all laboratory personnel and particularly women of childbearing age should be provided with information regarding immune competence and conditions that may predispose them to infection. Individuals having these conditions should be encouraged to self-identify to the institution's healthcare provider for appropriate counseling and guidance.

## Decontamination Methods

Decontamination in research laboratories is the process of removing or inactivating microorganisms to render surfaces, areas, devices, or equipment reasonably free from a risk of transmission of infectious agents or materials to laboratory personnel, the public, and the environment. In addition, it reduces or eliminates cross-contamination within the laboratory. Decontamination methods include sterilization (see Autoclave), disinfection (see "[Common Disinfectants for the Laboratory Guide Sheet](#)"), and antiseptics.

1. Sterilization. A procedure that kills all classes of microorganisms including viruses and bacterial and fungal spores. Sterilization is accomplished by heat, gases (ethylene oxide gas and hydrogen peroxide gas), radiation, plasma, and ozone.

A process is defined as "sterilization" if the probability of a microorganism surviving on an item subjected to the process/treatment is less than one in one million ( $10^6$ ); also known as the "sterility assurance level" ([BMBL](#)).

2. Disinfection. A physical or chemical means of killing nearly all pathogenic microorganisms including specific viruses, bacteria, and pathogenic fungi, but not bacterial spores. It is generally a less lethal means or process than sterilization. Its effectiveness depends on different factors including the nature and number of contaminating microorganisms, the amount of organic matter present, the types and condition of materials or instruments to be disinfected, and the prevailing temperature ([BMBL](#)).
3. Antisepsis. A means of inhibiting the growth and development of microorganisms without necessarily killing them. Antiseptic agents are usually formulated to be used on body surfaces such as skin or tissue.

Several important classes of chemical disinfectant are discussed in Table 7.1 (Reference - CDC/NIH: [Biosafety in Microbiological and Biomedical Laboratories 5th Edition](#): Disinfectants Comparison Chart-NIH Department of Education).

**Table 7.1 Chemical Disinfectants**

Property	Description
Disinfectant	<b>Bleach</b>
Effective Against	Bacteria spores; vegetative bacteria, most viruses (HIV, HBV, H1N1, MRSA), TB and fungi
Dilution/Shelf Life	1:10 (from 5% chlorine solution)/24 hours
Contact Time	Surfaces: 10 – 15 minutes; Liquid waste: 20 – 30 minutes
Usage	Spills; Liquid Waste; Equipment; Environmental Surfaces; BSC <sup>2</sup> ; etc.
Effects	Inactivated by organic matter; corrosive to eyes, skin, and respiratory irritant
Activity Level <sup>1</sup>	Intermediate
Disinfectant	<b>Ethanol</b>
Effective Against	Broad spectrum of bacteria and viruses; NOT effective against bacterial spores or norovirus
Dilution/Shelf Life	70% – 90%/stable
Contact Time	0.5 – 10 minutes <sup>3</sup>
Usage	Equipment; Surfaces; Skin antiseptic
Effects	Non-Corrosive, no residue, flammable (NO drain disposal), eye irritant
Activity Level <sup>1</sup>	Intermediate
Disinfectant	<b>Hydrogen Peroxide</b>
Effective Against	Broad spectrum of pathogens including H1N1, MRSA, norovirus, and parvovirus
Dilution/Shelf Life	3% – 6%/high to intermediate
Contact Time	1 – 10 minutes
Usage	Surfaces in BSL3; animal facilities; High throughput and other heavy equipment
Effects	Non-flammable, less corrosive than bleach
Activity Level <sup>1</sup>	High to Intermediate

1 - Based on Spaulding classification (BMBL-Appendix B) – Intermediate level: Kills vegetative microorganisms including *Mycobacterium tuberculosis*, all fungi, and inactivates most viruses. High level disinfectant: kills vegetative microorganisms and inactivates viruses but not high numbers of bacterial spores. Formulated to be used on medical devices, but not on environmental surfaces i.e. laboratory benches.

2 - Use of bleach in BSC or equipment must be followed by 70% ethanol swipe and/or sterile water swipe.

3 - Contact time varies for pure or mixed cultures.

## Sharps Precautions

One of the most common injuries in biomedical research is a needlestick injury. Effective engineering controls and work practices help minimize or eliminate the use of sharps or the risk of injuries from sharps. The following are considered sharps: needles, scalpels, blades, pipettes and broken glassware.

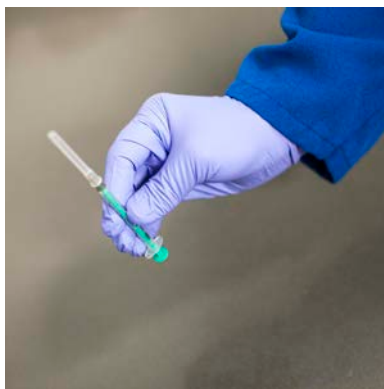
The following precautions must be followed when working with sharps.

1. Avoid or minimize the use of needles and other sharps whenever possible.
2. Use plastic labware over glass as a safer alternative.
3. Dispose of used needles immediately in a red puncture-resistant sharps container. Do not leave on benchtops or counter. Secure the lid when the sharps container is 2/3 full.
4. Wear appropriate PPE.
5. Needles MUST NEVER be recapped, removed from the syringe, sheared, bent or broken.
6. Never remove needle caps by mouth.
7. Always restrain or anesthetize animals prior to injection (i.p.; s.c.; or i.v.) to prevent inadvertent movement which may lead to needlestick injuries.
8. Use syringes with a Luer lock system to prevent the needle from detaching from the syringe during use.
9. Use a mechanical device to remove scalpel blades. NEVER remove them with your fingers.
10. Remove or sweep broken glass with a brush and dustpan, tongs, or forceps.

Use self-sheathing syringes whenever possible. If a needle must be recapped manually, use the One-Handed Scoop Technique:



Place cap on hard flat surface.  
Slide syringe needle into cap.



Scoop cap up with syringe needle  
so that cap is sitting on needle.



Using hard flat surface,  
press cap onto needle until  
cap snaps into place.

The Office of Environmental Health and Safety offers help in evaluating or selecting safety engineered needles. Contact [biosafety@usc.edu](mailto:biosafety@usc.edu) for any questions.

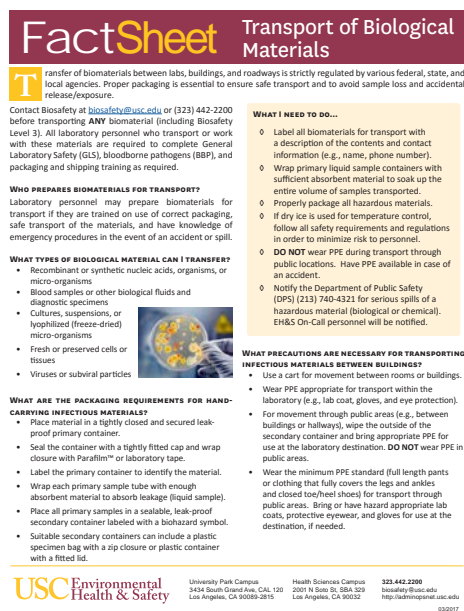
## Transport of Biohazards

Safe transport of biological materials between laboratories and between USC buildings is governed by university safety policy and best practices. Transport between USC campuses and safe shipment to domestic and international destinations are governed by DOT/IATA requirements and specialized training. Consult the [Transport of Biological Materials Fact Sheet](#) for more information (see Figure 7.5).

Biosafety practices include:

- Decontaminate the outside of the primary container before placing within the secondary container.
  - Decontaminate the secondary container before leaving the laboratory.
  - Wear minimum PPE (full length pants or clothing that fully covers the legs and ankles, closed-toe and heel shoes) in public corridors and outside the laboratory.
  - Within the laboratory, wear appropriate PPE as determined by risk assessment.
  - Have available biological spill clean-up supplies (disinfectant, absorbent materials). See [Biohazardous Spill Clean-Up Guide Sheet](#) for more information.
- Transport between Labs within a Building

Figure 7.5. Transport of Biological Materials Fact Sheet



<http://tiny.cc/usc-ehs-bioxprt>

The minimum requirements for safe transport of biological materials are:

- The sample must be tightly closed and secured in a leak-proof primary container.
- Each primary sample tube must be wrapped with absorbent material (if liquid), then placed in a sealable, leak-proof secondary container.
- The absorbent material must be sufficient to absorb the entire volume of the sample should leakage occur. Whenever feasible, avoid glass and use plastic during transport.

If glass vials or tubes must be used, wrap each tube individually with absorbent to cushion and absorb any liquid released from broken or damaged tubes.

- If dry ice is required, use an insulated outer (tertiary) container to contain the dry ice. The secondary container is placed within the tertiary container, and the dry ice is placed between the secondary and tertiary containers. **NOTE:** Dry ice is NEVER placed within a sealed container.
- The OSHA Bloodborne Pathogens Standard (1910.1030) requires that samples containing human blood or other potentially infectious materials (OPIM) must have a biohazard label on the outer container.
- Label the outer package with the agent name, the Principal Investigator (PI) name, and the PI contact information.

## Transport between Campus Buildings

The same standards for transport between laboratories apply for transport to laboratories between buildings. Additionally,

- Personnel must bring or have available at the destination the appropriate PPE (determined by risk assessment). Do NOT wear PPE in public areas, including elevators, stairways, and corridors.
- Use a cart to facilitate transport of samples between buildings.

## Transport on Public Roadways

There are specific restrictions and Department of Transportation (DOT) training requirements on the packaging and transport procedures for infectious materials on public roadways.

Contact [biosafety@usc.edu](mailto:biosafety@usc.edu) if biohazards need to be transported between UPC and HSC campuses.<sup>1</sup>

Self-transport of biohazards is limited to small quantities and is divided into two categories:

- Biohazards under “Materials of Trade” (MOT) exception can be self-transported in small quantities. **NOTE:** Small quantities as defined by the MOT exception are less than or equal to 0.5 kg or 0.5 L.
- Biohazards classified as “Category A” require additional training for personnel who transport and package these materials. Category A materials are biological substances in a form capable of causing permanent or life-threatening or fatal disease in otherwise healthy human or animals when exposure occurs.

Personnel who wish to transport or ship biological materials falling under the transport requirements for Infectious Substances regulated under this category must contact [biosafety@usc.edu](mailto:biosafety@usc.edu) for additional training and guidance.

For self-transport in a vehicle under the MOT exception:

- Use a packaging system which follows the packaging recommendations listed above for transfer between laboratories and buildings or packaging of equal or greater strength or integrity.

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<sup>1</sup> Do NOT use public trams, shuttles, or taxi transport for transport of biohazards between campuses or on public roads.

- Use the original packaging provided for the biological materials whenever possible.
- Label primary and outer packaging with the proper shipping name of the material, and the name and phone number of the laboratory sender and recipient contact information.
- If used, dry ice must be placed outside of the secondary container within the tertiary container, not inside a closed, non-venting container. The outside package must be marked with the words, "dry ice."
- If transported by private vehicle, the vehicle used must be for direct and exclusive transport of the sample to the destination (no other stops permitted).
- The outer packaging must be securely closed and secured against shifting or movement between packages during transport.

#### References

- 29 CFR 1910.1030. Bloodborne Pathogens Standard
- 29 CFR Part 1910.1200, Hazard Communication
- 29 CFR Part 1910.1450. Occupational Exposure to Hazardous Materials in Laboratories
- 40 CFR Part 262, Standard Applicable to Generators of Hazardous Waste
- 49 CFR Parts 171-180, Hazardous Materials Regulations 171.1 (d), Ground transport of hazardous materials; 173.6, Materials of trade exceptions.

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## 8.0 Laboratory Equipment

### Biosafety Cabinets

Biosafety cabinets (BSCs) are the primary and most effective laboratory devices for working safely with infectious microorganisms. They are designed to contain infectious agents as well as splashes and aerosols released during work within the cabinet. Proper use of the biosafety cabinet is detailed in EH&S' [Biosafety Cabinets Fact Sheet](#) (see Figure 8.1). BSCs provide three basic types of protection:

1. Personnel protection from exposure to splashes and aerosolized infectious agents inside the cabinet.
2. Experimental products protection from contamination from infectious materials inside and outside the cabinet.
3. Environmental protection from potential release of infectious agents.

#### Biosafety Cabinet (BSC) Classes

Biosafety cabinets fall into the following classes: Class I, II, and III. At USC, the Class II BSC is commonly used in the laboratory for protection of the user and to keep the research products free from contamination. A Class II vertical laminar-flow BSC is an open-front, ventilated cabinet designed to allow only sterile air from a HEPA-filtered supply into the cabinet and over the work surface. Half of the supply HEPA-filtered air flows downward to the front exhaust grill, the other half to the rear exhaust grill. There are four types of Class II cabinets: A1, A2, B1, and B2 (see Table 8.1).

Figure 8.1. Biosafety Cabinets Fact Sheet

### FactSheet

**A** Class II biosafety cabinet (BSC), sometimes referred to as a tissue or cell culture hood, is an engineering control used in biological research that provides primary containment from infectious microorganisms. Used properly, a BSC protects the worker, the environment, and the material that is being used.

**HOW DOES A BIOSAFETY CABINET WORK?**  
The BSC uses High Efficiency Particulate Air (HEPA) filtered vertical laminar airflow to create a barrier to airborne particles, such as microorganisms. The sheet of filtered air protects the material inside the cabinet.

**WHEN SHOULD I USE A BIOSAFETY CABINET?**  
BSCs are always required for manipulations of airborne transmitted pathogens (e.g., influenza virus), biological materials that are likely to create aerosols, and when working with high concentrations of infectious agents.

**HOW DO I USE A BIOSAFETY CABINET PROPERLY?**

- Set up the workflow from "clean to dirty."
- Limit movement of "dirty" items over "clean" ones.
- Place materials 3-5 inches away from front and back grills; set sash at 8-10 inches above base.
- Minimize hand movements inside the cabinet.

**HOW DO I CLEAN AND DISINFECT A BIOSAFETY CABINET?**

- Use 10% bleach for a contact time of 10 minutes.
- Wipe up bleach with water since it will oxidize stainless steel and cause pitting.
- Use a final rinse of 70% EtOH to remove the water and any residual bleach.

### Biosafety Cabinet Class II


**WHAT I NEED TO KNOW**

- Use a risk assessment to determine if a BSC is required. Contact the IBC or Biosafety Office for assistance.
- Blocking the front and back grills and/or creating turbulence (loading the cabinet with too many items or moving hands around or in and out of the cabinet) diminishes the efficiency of the cabinet.
- ALL BSCs must be decontaminated prior to being moved. No exceptions.

**MAY I USE OPEN FLAMES IN A BIOSAFETY CABINET?**  
**NEVER use an open flame in a BSC.** It disrupts air flow, compromising protection of both the worker and the work. An open flame may ignite flammable liquids and materials close by. Alternative methods such as electric incinerators or disposable inoculating loops are recommended.

**DO ULTRAVIOLET LIGHTS PROVIDE DECONTAMINATION IN A BIOSAFETY CABINET?**  
Ultraviolet (UV) light is an extra precaution in keeping the work area decontaminated between uses. To optimize its effectiveness make sure you clean, maintain, and monitor it periodically. Running a BSC for 30 minutes before use may be as effective as keeping a UV light on during periods of non-use.

**WHY IS BSC CERTIFICATION REQUIRED?**  
BSCs must be tested and certified on site to ensure that they are providing adequate personnel and environmental protection. BSCs are certified: (a) at the time of installation, (b) at least annually thereafter, and (c) at any time the BSC is moved.



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<http://tiny.cc/usc-bscII-fs>

#### Class II Type A

- Class II, Type A1 (formerly A) BSC vents 30% directly into the laboratory (70% recirculated in the BSC) and cannot be used for work involving volatile toxic chemicals.
  - 75 FPM (feet per minute)
  - May have biologically contaminated positive pressure plenum.
- Class II, Type A2 (formerly B3; see Figure 8.2) BSC may be a standalone unit or ducted to the outside through the building exhaust system designed for hazardous substances. It is suitable for work with infectious agents and minute amounts of hazardous chemicals.

**Table 8.1. Comparison of BSC Characteristics (adopted after BMBL Appendix A)**

BSC Class	Face Velocity (FPM)	Air Flow Pattern	Applications <sup>1</sup>		Protection Provided	BSL/Hazard Corrected
			NVTC&R <sup>2</sup>	VTC&R <sup>3</sup>		
II A1	75	70% recirculated thru HEPA; 30% exhausted to room or outside thru HEPA.	Yes; small amounts	No	Personnel Environment Product	1 – 3/ Aerosols, splashes, and spatters
II B1	100	30% recirculated thru HEPA; 70% exhausted thru dedicated duct to outside thru HEPA	Yes	Yes; sml amts <sup>4,5</sup>	Personnel Environment Product	1 – 3/ Aerosols, splashes, and spatters
II A2	100	Similar to II A1, but has 100 FPM and plenums under negative pressure; ducted exhaust air	Yes	When exhausted to outside; sml amts <sup>4,5</sup>	Personnel Environment Product	1 – 3/ Aerosols, splashes, and spatters
II B2	100	No recirculation; 100% exhaust to outside thru HEPA filter	Yes	Yes; sml amts <sup>4,5</sup>	Personnel Environment Product	1 – 3/ Aerosols, splashes, and spatters
III	N/A	HEPA-filtered supply air; ducted exhaust air passes thru 2 HEPA filters in series	Yes	Yes; sml amts <sup>4,5</sup>	Maximum for Personnel Environment Product	3 – 4/ Aerosols, splashes, and spatters

1 - While HEPA filters are effective at trapping particulates and infectious agents, they do not capture radionuclides, volatile toxic chemicals, and gases.

2 - Non-volatile toxic chemicals and radionuclides.

3 - Volatile toxic chemicals and radionuclides.

4 - Special installation venting and/or filters may be needed. Do not discharge into a room if volatile chemicals are used.

5 - Keep concentrations below the lower explosion limit.

### Class II Type B

- In Class II, Type B1, 70% of air downflow exits through the rear grill and HEPA-filtered before being discharged from the building. The remaining 30% is drawn through the front grill and HEPA-filtered through the supply filter before being re-circulated. Class II B1 must be ducted to a dedicated exhaust system rated for hazardous substances. It is suitable for work with infectious agents treated with minute amounts of hazardous chemicals.
- In Class II, Type B2, 100% of air downflow is exhausted outside the building through a dedicated duct (see Figure 8.3). An interlock system prevents the supply blower from operating when the exhaust flow is insufficient. Class II B2 is suitable for infectious agents treated with hazardous chemicals and radionuclides.

### Class III Biosafety Cabinet

The Class III BSC provides the highest level of protection and is suitable for work with infectious agents in BSL3 and BSL4 laboratories. It is completely enclosed and access to the work surface is only by means of heavy duty rubber gloves attached to ports in the cabinets. The cabinet is maintained under negative pressure with HEPA-filtered supply air and double HEPA-filtered exhaust air.

Figure 8.2. Class II, Type A2 biosafety cabinet

#### BIOSAFETY CABINET (BSC) CLASS II, TYPE A2

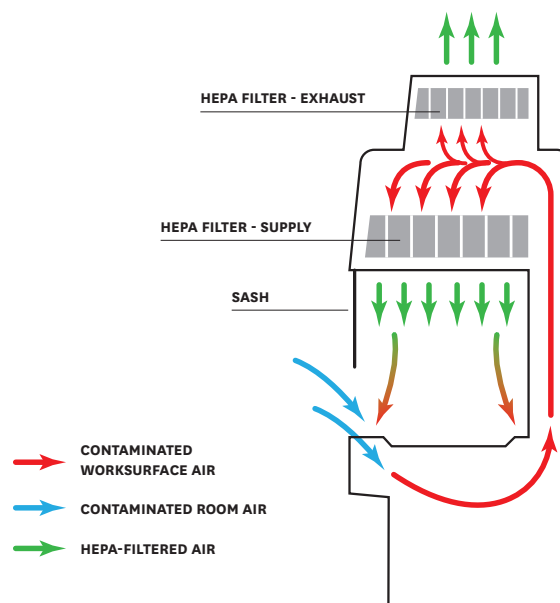
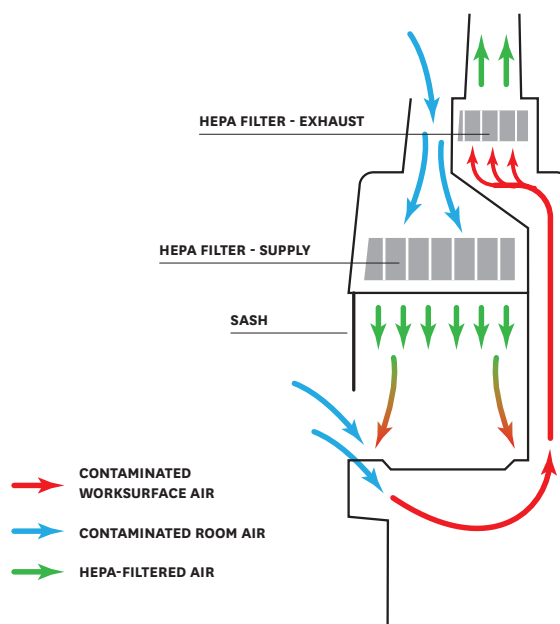


Figure 8.3. Class II, Type B2 biosafety cabinet

#### BIOSAFETY CABINET (BSC) CLASS II, TYPE B2



## Biosafety Cabinet Set-Up and Use

Prior to using a BSC:

- Confirm that pressure gauges, flow indicators, and alarms are within recommended posted ranges. Discontinue any planned work if the airflow is outside range.
- Protect the vacuum line by connecting suction flasks to an overflow collection flask containing the appropriate disinfectant, and to an in-line HEPA filter.
- Ultraviolet lights are not required in the BSCs.
- Turn on the blower and fluorescent lights at least 10 minutes prior to work; ensure the drain valve is closed.
- Disinfect the cabinet work surface and interior walls with 1:100 dilution of household bleach (i.e., 0.05% sodium hypochlorite), rinse with sterile water to remove bleach, and wipe with 70% ethanol (EtOH) to remove water.
- Load only the items needed for the planned procedure.
- Materials can be wiped with disinfectant before placing them inside the BSC.
- Place materials at least six inches from the front grill.
- Equipment (e.g., centrifuge, blender) that creates air turbulence must be placed in the back of the cabinet and other work must stop while the equipment is running.
- Work as far to the back of the BSC workspace as possible.
- Segregate contaminated and clean items by working from "Clean to Dirty" (or sterile to contaminated).
- Clean-up spills immediately according to the laboratory SOP.

Figure 8.4. Safe Alternatives to Open Flames Fact Sheet

**FactSheet** Safe Alternatives to Open Flames in a BSC

Use of an open flame (Bunsen burner) within a biological safety cabinet (BSC) directly impacts the BSC's laminar air flow and compromises its efficiency and user safety. Significant damage from lab fires has occurred at USC in the past that were directly attributed to use of open flames in BSCs.

The World Health Organization's (WHO) Laboratory Biosafety Manual, NSF/ANSI Standard 49, BSC manufacturers, and academic institutions STRONGLY discourage/forbid the use of Bunsen burners inside the BSC for the following reasons:

- **Air flow disruption.** Heated air from an open flame mixes with the Class II BSC's downward laminar flow to produce turbulence and recirculation inside the work area. Aerosolized particles will be distributed throughout the BSC, compromising both personnel and product protection.
- **Excessive heat build-up.** This can damage the HEPA filter and housing resulting in loss of BSC integrity and migration of contaminants into and out of the BSC. Additionally, loss of media components (e.g., growth factors, amino-acids, and vitamins) and heat-sensitive cell lines as well as worker discomfort impact research stability and quality.
- **Fire or explosion hazard.** Excessive heat may cause flammable liquids used for disinfection to vaporize creating a flammable atmosphere. An open flame or electrical discharge may ignite the vapor causing a flash fire or explosion.
- **Void Manufacturer's Warranty and UL Listing.** Fires within a BSC caused by use of open flames will nullify the manufacturer's warranty and UL listing.

**SAFE ALTERNATIVES TO BUNSEN BURNERS**

- Use disposable sterile loops and sterile lab supplies. This eliminates the need to use open flames for sterilizing.
- Autoclave utensils and equipment prior to use. Place loops, spreaders, needles, forceps, scalpels and other tools in autoclavable plastic or wrap in autoclavable foil.

**WHAT I NEED TO KNOW**

- Do not use an open flame in the BSC. The BSC provides a near microbe-free environment.
- Heated air in the BSC disrupts laminar flow and impacts the HEPA filter/housing, media, cell lines, and workers.
- Questions? Email [biosafety@usc.edu](mailto:biosafety@usc.edu).
- Use a Bacti-Cinerator to sterilize loops and needles safely and conveniently while preventing infectious spatter and cross-contamination.
- The Electrical Bunsen Burner combines the efficiency of a gas burner with the safety and control of an electric heater. It is ideal for sterilizing inoculating needles and loops, and for heating small flasks, test tubes, and beakers.
- The Bead Sterilizer provides a safe, effective, and convenient method for sterilizing small instruments without using flames, gases, or chemicals.

**REFERENCES**

- Biosafety in Microbiological and Biomedical Laboratories (BMBL) 5th edition.
- WHO Biosafety Manual 3rd Edition, WHO 2004
- Sterile-Disposable Loops (Pioneer Scientific catalog #22-363-596)
- Bacti-Cinerator (VWR catalog #101412-622)
- Electrical Bunsen Burner (Daigler Scientific catalog #EF3540)
- Instrument Micro Bead Sterilizer (Kent Scientific Corporation)

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- During work inside the BSC:
  - Do NOT block the front or rear intake grill.
  - Do NOT use a Bunsen burner. (see Figure 8.4 [Safe Alternatives to Open Flames in a BSC Fact Sheet](#)).
  - Keep work surfaces clutter-free.
  - Avoid rapid movement of arms and hands or frequent in and out motions. Do NOT rotate hands or arms out of the work area.
  - Do NOT place non-sterile materials upstream of sterile materials.
- After work, wipe down the exterior of all materials with the appropriate disinfectant (see [Section 7 Decontamination Methods](#)) prior to removing from the BSC.
- Decontaminate and properly dispose of any generated biological waste (see [Section 9 Biohazardous Waste Management](#)).

- Disinfect all interior work surfaces and walls with the appropriate disinfectant followed by a 70% ethanol wipe-down.
- Surfaces under the work grills must be periodically decontaminated.

Inspection of the cabinet must include tests for the cabinet integrity, downflow velocity profile, air flow smoke pattern, HEPA filter leaks, ventilation rate and negative pressure, alarms, and interlocks, if applicable.

### BSC Certification

All USC BSCs must be tested and certified: after installation; when moved or repaired; and annually by an accredited certifier company (see [USC-approved vendors](#)) in accordance to NSF/ANSI 49.

### BSC Decontamination

BSCs must be decontaminated by a qualified professional before conducting the following: general maintenance and filter changes; repairs; and relocation.

## Autoclave

Autoclaving is a sterilization process that uses pressurized steam to kill microorganisms. It is the preferred method for decontamination of regulated medical waste containing bacteria, viruses, and other biological waste, and sterilization of laboratory equipment, materials, media and reagents (see Figure 8.5).

### Autoclave Cycle

Steam sterilization is a function of pressure, temperature, and time. The process employs saturated steam under a pressure of 15 psi to achieve a chamber temperature of at least 121°C (250°F) for a minimum of 15 minutes. Increased cycle time may be necessary for some load types and volume.

The autoclave cycle settings will depend on the nature of the load. Gravity Cycle must be used for wrapped dry materials such as sharps (scalpels, scissors), materials in containers, pipette tip boxes, glassware, and biological waste, and Liquids Cycle for liquid media, water, or liquid biological waste. Table 8.2 summarizes the general guidelines for sterilization of lab equipment, media, and biohazardous waste.

**Figure 8.5. Large chamber autoclave**



Source: Priorclave

**Table 8.2. Autoclaving guidelines for sterilization of lab equipment, media, and bio waste.**

Items/ Materials	Cycle	Preparation	Loading <sup>1</sup>	T/psi	Duration <sup>2</sup>	Exhaust
Glassware	Gravity	<ul style="list-style-type: none"> <li>Wash/rinse/dry and wrap with butter paper or autoclaving foil</li> <li>Cover open ends with cotton or other steam-penetrable plug</li> </ul>	Place in rigid secondary container	121°C (250°F)/ 15 psi	20 – 30 minutes	Clean: Fast/dry Dirty: Slow
Liquid	Liquid	<ul style="list-style-type: none"> <li>Fill containers no more than 50% capacity</li> <li>Ensure caps are loosened or use vented closure</li> </ul>	Place upright in secondary container (pan)	121°C (250°F)/ 15 psi	30 – 60 minutes	Slow
Dry (wrapped)	Gravity	<ul style="list-style-type: none"> <li>Wash/rinse/dry and wrap with autoclaving foil</li> </ul>	Place in rigid secondary container	121°C (250°F)/ 15 psi	30 – 60 minutes	Fast/dry
Bio red bag	Gravity	<ul style="list-style-type: none"> <li>Position biohazard bags on their sides</li> <li>Close bags loosely to allow steam penetration</li> </ul>	Place bags inside rigid plastic containers to catch spills	121°C (250°F)/ 15 psi	60 minutes	Slow

1 - Materials must be placed in the center of the autoclave whenever possible.

2 - Treatment or processing times will vary according to load size: The larger the load, the longer the decontamination time.

### Preparation of Materials

Adequate steam penetration is essential for effective decontamination; do not compact or overfill biohazardous waste bags.

When using an autoclave, place autoclave loads in large, leak-proof, and autoclavable polypropylene or stainless steel pans during autoclaving to catch spills. Biohazardous bags must be positioned on their sides with the neck loosely closed or opened to allow steam penetration.

Water may be added to the bag to facilitate heat transfer during the decontamination process.

When loading the autoclave, create space between the items to allow steam circulation. For liquid cycle autoclaving, the liquid containers must be no more than half filled with the caps loosened or fitted with a cotton plug (or other steam-penetrable and autoclavable plug), and placed in shallow secondary containers (e.g., pan).

## Basic Operation

- Always follow the manufacturer's operating instructions.
- Don appropriate PPE including lab coat, heat resistant gloves, and eye protection when loading and unloading the autoclave.
- Place the items to be autoclaved in the chamber.
- Close and seal the autoclave door.
- On the autoclave keypad, select the appropriate:
  - Cycle of the load/material: Gravity, Liquid, or Dry.
  - Sterilization time. Note that the actual time will be longer (additional 12 – 15 minutes) to adjust for temperature and pressure ramping inside the chamber.
  - The sterilization temperature is 121°C.
  - Alternatively, a pre-programmed cycle may be selected.
- Start the autoclave cycle and fill out the autoclave log.
- At the completion of the cycle, wait until the pressure drops to zero and the temperature is below 121°C before opening the door.
- Don appropriate PPE (e.g., lab coat, heat-resistant gloves, closed-toed shoes, safety glasses) and open door slowly and only slightly to allow steam to escape. Avoid standing directly in front of the autoclave door.
- Keep ajar for 5 – 10 minutes before opening fully.
- Check autoclave tape indicator for a color change and print-out from the recorder to see if time and temperature were attained.
- When unloading liquids, avoid agitating the containers or removing caps.

- Place autoclaved biohazardous waste bags in the appropriate biohazardous bins.
- Shut the autoclave door.

## Monitoring

### Chemical Indicators

Chemical indicators provide quick visual cues of heat penetration following normal autoclave cycles. However, they are not proof that microorganisms were killed during decontamination.

- Tape Indicators. Tape indicators (see Figure 8.6) are adhesive-backed paper tape with heat sensitive markings (e.g., diagonal stripes and/or the word “sterile”) that appear only when the tape is exposed to normal autoclave decontamination temperatures of 121°C (250°F).

**Figure 8.6. Autoclave tape indicator**



Source: Hu-Friedy

**Figure 8.7. Chemical indicator strips**



Source: barberdts

- Integrated Chemical Indicator Strips. Strips (see Figure 8.7) will display a color change after exposure to normal autoclave operating temperatures of 121°C (250°F) for several minutes. Do not use autoclave tape as the sole indicator of sterilization.

### Biological Indicators

Biological indicators reveal that microorganisms are killed during the decontamination cycle. There are three types of biological indicators: *Geobacillus stearothermophilus*, *Bacillus subtilis*, and *Clostridium sporogenes* spores. *G. stearothermophilus* is commonly used to validate microorganism death after steam autoclaving sterilization (also used in Vapor-Phase-Hydrogen-Peroxide sterilization validation).

Test biological indicator vials (see Figure 8.8) are placed among loads and autoclaved; control biological indicator vials are not autoclaved and kept at room temperature.

After removal from the autoclave, the test vials are incubated at 55 – 60° C along with the control vials for up to three days.

Clear test vials with no evidence of growth or turbidity indicates successful decontamination. The control vials shall exhibit growth and turbidity. If the autoclaved vials exhibit evidence of growth, the test must be repeated. If a second failure occurs, notify EH&S and place a “DO NOT USE” sign on the autoclave.

**Figure 8.8. Bio indicator**



Source: hygitech

### Autoclave Log

Obtain an autoclave log book and place near the autoclave. Create an entry log to include the following information:

- Name and contact information of the operator/autoclave user
- Date and time used
- Process type
- Duration of the run (cycle time)
- Type of material processed
- Chemicals or biological indicator results
- Sterilization temperature, pressure, and length of time
- Malfunction/Maintenance or repair

### Autoclave Safety

#### Potential Risks

There are potential risks associated with an autoclave that are linked to how it operates. These include:

- Burns from residual steam
- Boiling liquid and spillage in the autoclave
- Hands and arms injury from contact with autoclave door
- Injuries from pressure release/explosion

### Preventative Measures

- Always wear lab coat, eye protection, heat-resistant gloves, and closed-toe shoes when operating the autoclave.
- Do not attempt to open the door while the autoclave is running.
- Always wait for the pressure to reach zero before attempting to open the autoclave.
- Avoid standing directly in front of the door.
- Avoid superheating liquids.
- If problems occur during the cycle, abort the cycle and report it to the PI, Lab Manager, and/or other responsible person.

The following items (or waste) are not recommended for autoclaving:

- Radioactive materials
- Pathological waste
- Hazardous chemicals (flammable, corrosive, toxic, reactive)
- Household bleach
- Liquid in a sealed container
- Toxins and prions

## Centrifuges

(Adapted from [OSHA QuickFacts Centrifuges](#))

Centrifuges (see Figure 8.9), which operate at high speed, have great potential for injuring users if not operated properly. Unbalanced centrifuge rotors can result in injury or death. Sample container breakage can release aerosols that are harmful if inhaled.

The majority of all centrifuge accidents result from user error. To avoid injury, workers should follow the manufacturer's operating instructions for each make and model of centrifuge that they use.

Follow these steps for the safe operation of centrifuges:

- Do not use a centrifuge that has debris inside or material splashed on the sides.
- Ensure that centrifuge bowls and tubes are dry.
- Ensure that the spindle is clean.
- Use matched sets of tubes, buckets and other equipment.
- Always use safety centrifuge cups to contain potential spills and prevent aerosols.

**Figure 8.9. Table top centrifuge**



Source: Scilogex

- Inspect tubes or containers for cracks or flaws before using them. DO NOT use any cracked or damaged tubes or containers.
- Avoid overfilling tubes or other containers (e.g., in fixed angle rotors, centrifugal force may drive the solution up the side of the tube or container wall).
- Ensure that the rotor is properly seated on the drive shaft.
- Make sure that tubes or containers are properly balanced in the rotor.
- Only check O-rings on the rotor if you are properly trained. Otherwise you may harm the rotor.
- Apply vacuum grease in accord with the manufacturer's guidelines.
- Do not exceed the rotor's maximum run speed.
- Close the centrifuge lid during operation.
- Make sure that the centrifuge is operating normally before leaving the area.
- Make sure that the rotor has come to a complete stop before opening the lid.

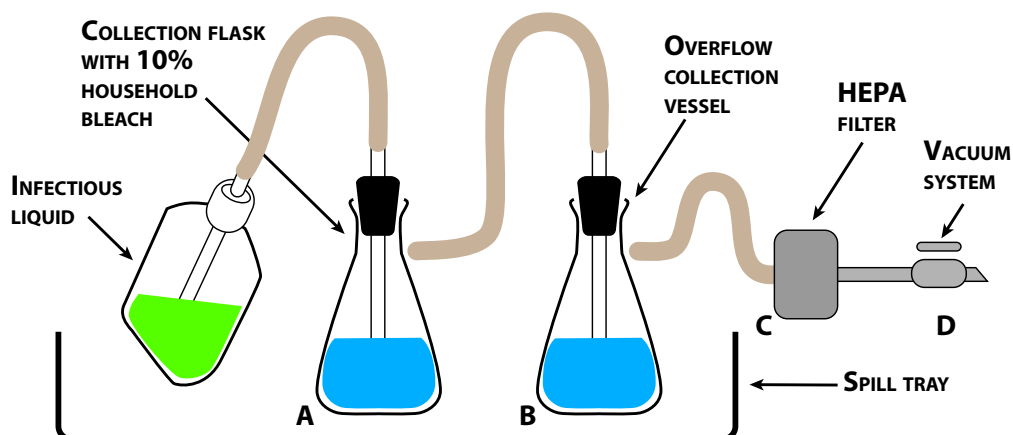
When centrifuging infectious materials, wait 10 minutes after the rotor comes to a complete stop before opening the lid. Open the rotor lid only in the BSC. If a spill has occurred, use appropriate decontamination and cleanup procedures for the spilled materials. Report all accidents to your supervisor immediately.

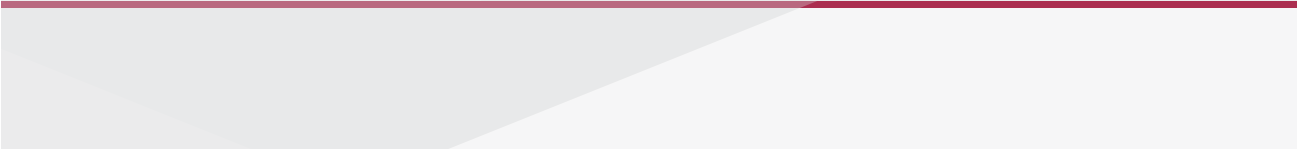
## Inline-Vacuum Traps

(adapted from BMBL)

Aspirator bottles or suction flasks should be connected to an overflow collection flask containing appropriate disinfectant, and to an in-line HEPA or equivalent filter (see Figure 8.10). This combination will provide protection to the central building vacuum system or vacuum pump, as well as to the personnel who service this equipment. Inactivation of aspirated materials can be accomplished by placing sufficient chemical decontamination solution into the flask to inactivate the microorganisms as they are collected. Once inactivation occurs, liquid materials can be disposed of as noninfectious waste.

**Figure 8.10. In-line Vacuum Trap**





Protection of vacuum systems must be addressed. Figure 8.10 illustrates one method to protect a house vacuum system during aspiration of infectious fluids.

The left suction flask (A) is used to collect the contaminated fluids into a suitable decontamination solution; the right flask (B) serves as a fluid overflow collection vessel. An in-line HEPA filter (C) is used to protect the vacuum system (D) from aerosolized microorganisms.

Other devices, such as Guardian canisters or other inline plastic disposable suction canisters may be used.

## **Tissue-Disrupting Devices**

There are several devices used for the disruption of tissue. They include sonicators, vortexes, bead beaters, douncers, tissue grinders, and mortar and pestle. The use of any of these devices may cause potentially infectious aerosols. Therefore, a risk assessment should be undertaken by the user to determine if the equipment requires the use of a BSC or another form of physical containment to prevent exposure to infectious aerosols.

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## 9.0 Biohazardous Waste Management

### Biohazardous Waste

Biohazardous waste is a term used to define different waste streams that contain biological hazards. It is defined by the California Department of Public Health as including the following:

- Medical waste
- Clinical waste
- Biomedical waste
- Laboratory waste

For complete definitions of biohazardous waste streams, refer to the [Medical Waste Management Act January 2017](#), California Health and Safety Code, Sections 117600 – 118360.

Solid biohazardous waste is managed per the following:

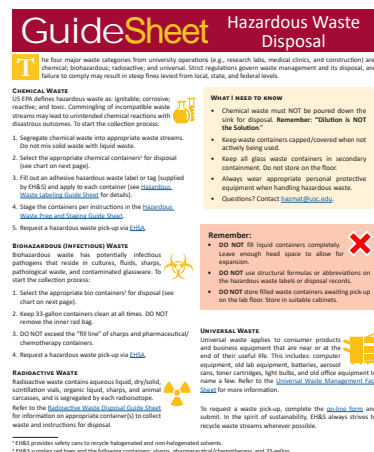
- Use red biohazard bags marked with the international biohazard symbol to contain solid waste.
- Place red biohazard bags in a rigid, leak-proof container with a tight-fitting lid (see Figure 9.1). Ensure that these containers are also marked with the international biohazard symbol.
- Secure the lid on the container unless it is in use.
- Do NOT overfill the container. The container lid must fit tightly to ensure a good seal.

Consult the [Hazardous Waste Disposal Fact Sheet](#) (see Figure 9.2) for general biowaste disposal guidelines.

Figure 9.1. Bio red bag and container



Figure 9.2. Hazardous Waste Disposal Fact Sheet



Biohazardous liquid waste is: liquid media, human serum, and any other liquid that is potentially infectious. Liquid biohazardous waste may be treated with a 1:10 dilution of normal household bleach, left for 30 minutes, then disposed to the sanitary sewer followed by running water until the bleach odor is less perceptible.

Bleach is not effective when there is protein present, therefore, spraying bleach on solid material such as animal bedding or feces is not an effective decontamination method. Solid materials such as bedding and feces are disposed of as solid biohazardous waste.

### Sharps

A sharp is any object or device that has a rigid corner and/or sharp edge and that can cut or pierce tissue.

These objects or devices may cause injury to employees if not handled carefully. They can be especially dangerous if contaminated with biohazards. Examples of sharps include: needles; scalpels; blades; broken glass; glass slides; glass Pasteur pipettes; or anything that can puncture the skin.

Follow the steps below to dispose of sharps properly:

- Place all sharps in a red sharps container (see Figure 9.3) per the Medical Waste Management Act. Sharps containers are supplied by EH&S and are pre-labeled with the international biohazard sign.
- Never fill sharps container above the "No Fill Line" on the container.
- Close the lid once the container is 2/3 full.
- Request a waste pick up via [EHSA](#) or call EH&S at 323-442-2200.

**Figure 9.3. Sharps containers**



### Pharmaceuticals

Pharmaceuticals are defined as over-the-counter or prescription human or veterinary drugs. Follow steps below for proper disposal of pharmaceuticals (expired or no longer needed).

#### General Pharmaceuticals

General pharmaceuticals not categorized as controlled substances may be disposed of per the following:

- Place in a pre-labeled pharmaceutical container (see Figure 9.4) provided by EH&S.
- Never fill the pharmaceutical container above the "No Fill Line".
- Close the lid once the container is two thirds full.
- Request a waste pick up via [EHSA](#) or call EH&S at 323-442-2200.

**Figure 9.4. Pharmaceutical container**



Source: MET

### **Controlled Substances**

Consult the [Controlled Substance and Precursor Chemical Program](#) web page for information on waste management and disposal. For more information, contact [ehs-cs@usc.edu](mailto:ehs-cs@usc.edu).

### **Pathological Waste**

Pathological waste is not addressed in the Medical Waste Management Act January 2017, California Health and Safety Code, Sections 117600 – 118360. At USC, the pathological waste stream consists of the following:

- Human organs, tissues, body parts, and fluids
- Human/animal tissues or other materials treated with a pathogen that are potentially infectious
- Animal carcasses of animals that have been treated with a pathogen

These materials are placed in a white pathological waste bucket (see Figure 9.5) which is placed in a freezer for pickup by EH&S waste technicians.

**Figure 9.5. Pathological waste container**



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## 10.0 Biohazard Use Authorization

The iStar system is a web-based application routing and tracking system that increases the efficiency of the approval and administrative processes for projects and protocols involving human subjects and animals in research. It has replaced a cumbersome and paper-intensive process. iStar is a collaborative effort between the University of Southern California and the Children's Hospital Los Angeles (CHLA) to standardize and computerize the Institutional Review Boards (IRBs) at USC and CHLA. In 2013 and 2014, the IACUC, Biosafety and Radiation Safety Committees have also utilized the iStar system for their respective submission processes.

### IBC Registration Process

Principal Investigators (PIs) must have an approved Biohazard Use Authorization (BUA) for research studies involving:

- Recombinant or synthetic nucleic acids
- Human and non-human primate (NHP) blood
- Human, animal, and plant pathogens
- Other potentially infectious human or non-human primate materials (OPIM) e.g., cells or tissue
- Specific hazardous chemicals used in biomedical research e.g., chemotherapy, carcinogens, and toxins of biological origin.

Three items are ultimately required to conduct research with biohazards:

- Complete and submit a BUA application to the IBC for review through iStar.
- Ensure that staff on the project have undergone the appropriate training.
- Undergo the annual biosafety inspection.

### Registering a BUA

All research projects using biohazards must be registered with IBC by completing the Biohazard Use Authorization (BUA) on [iStar](#). For instructions and a guide to the [iStar](#) registration and submission process, visit iStar. Renewals or amendments to existing approved BUAs must be completed using the [iStar](#) system. The BUA must be renewed every 3 years.

Amendments are required for the following:

- Location changes
- Personnel update
- Addition of new biohazards
- Changes in research scope
- Addition of animal(s) to BUA

If animals are used in the research project, the Institutional Animal Care and Use Committee (IACUC) and IBC submission can take place concurrently. However, final approval by the IACUC is contingent upon review and approval of BUA by the IBC.

Experiments using recombinant DNA and synthetic nucleic acids are covered by the *NIH Guidelines for Recombinant and Synthetic Nucleic Acid Research*. Projects that have a major impact on human health must be reviewed at the highest levels by the NIH Director and the Recombinant Advisory Committee (RAC) as well as the local IBC (see Table 10.1). Any project involving transferring nucleic acids into humans must also be reviewed by the local IRB.

**Table 10.1. NIH or OBA Review**

Review & Approval Entities	Experiment	Section
IBC, NIH	Transfer of drug resistance that affects disease control	III-A
IBC, NIH	Cloning toxin molecules with LD50 <100 ng/kg body weight	III-B
IBC, IRB	Transfer of recombinant or synthetic nucleic acid into human subjects	III-C

Other experiments must be reviewed by the USC IBC prior to or concurrent with experiment initiation. This is outlined in Table 10.2.

**Table 10.2. USC IBC Review**

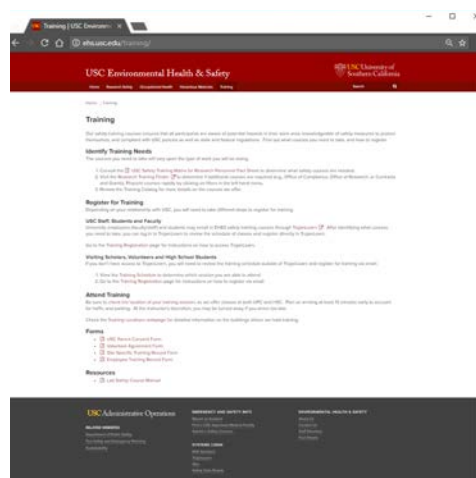
Review & Approval Entities	Experiment	Section
IBC approval prior to initiation	<ul style="list-style-type: none"> <li>Many experiments involving whole animals</li> <li>Cloning or host-vector experiments using recombinant and synthetic nucleic acid or microorganisms in Risk Group 2, 3, 4, or restricted agents</li> <li>More than 10 L of culture in one vessel</li> </ul>	III-D
IBC notification concurrent with experiment initiation	<ul style="list-style-type: none"> <li>Creation of transgenic rodents (genome altered by rDNA introduced into germline) that require BSL-1 containment</li> <li>The formation of recombinant nucleic acid molecules containing no more than two thirds of the genome of any eukaryotic virus propagated in tissue culture</li> <li>Recombinant or synthetic nucleic acid modified whole plants except those under Sections III A, B, C, and D</li> </ul>	III-E
IBC registration required	<ul style="list-style-type: none"> <li>Recombinant or synthetic nucleic acids not in microorganisms or viruses</li> <li>Purchase of transgenic rodents that require BSL-1 containment</li> <li>E. coli K12 host-vector system</li> </ul>	III-F

## Training

Prior to IBC approval of the submitted BUA, all personnel who are listed on the BUA must have undergone appropriate training. Biosafety training currently offered at USC includes bloodborne pathogen training (BBP), principles of biosafety training (BIO), viral vector training (VVT), and shipment of biohazardous materials (BHS) training.

Initial and annual refresher training is required. Information and registration for safety training is available at [Research Safety Training](http://ehs.usc.edu/training) (see Figure 10.1). All training must be documented through [EHSA](http://ehs.usc.edu/training).

**Figure 10.1 | EH&S Training Website**



<http://ehs.usc.edu/training>

## DAR/Biosafety Meetings

Research groups whose projects involve biohazards in animals must attend a DAR/Biosafety meeting prior to initiating any work. The meetings are mandated by the IBC, noted in the BUA approval, and scheduled through the Biosafety Program. USC veterinarians and Biosafety Team members will meet with research groups to review all biosafety and animal welfare aspects of the projects. Contact [biosafety@usc.edu](mailto:biosafety@usc.edu) to schedule a meeting.

## Occupational Medicine Program for Researchers Listed on the BUA

During review of BUAs in iStar, IBC members or the BSPs identify any occupational medical requirements for the project participants. If animals are used in the research project, participation in the [Animal Exposure Program](#) is required and there may be other recommendations for researchers, DAR staff, veterinarians, and visitors to the vivaria.


## Inspections

As a condition of BUA approval/re-approval, the IBC requires annual inspections of all laboratory spaces listed in the BUA. The inspection focuses primarily on engineering controls, administrative controls and PPE.

The following are biosafety requirements for the laboratory.

1. Biosafety training records must be documented and readily available.
2. [Pathogen Data Sheets](#) (PDS; see Figure 10.2), BBP Exposure Control Plan (ECP), and Chemical [Safety Data Sheets](#) (SDS) must be available for review by lab members.
3. Door signs must be posted on the entrance doors with the following information:
  - a. Universal biohazard symbol
  - b. The word "Caution"
  - c. The overall biosafety level
  - d. The PI's contact information (name, telephone number) in case of emergency
4. Biological hazard warning labels (see [Figure 12.9](#)) with the universal biohazard must be affixed to laboratory equipment (e.g., freezers, refrigerators, incubators, BSC, centrifuges, biohazard waste containers, and water baths). The labels must be readily visible and used to identify equipment used to store, manipulate, or dispose of infectious materials.

Figure 10.2 | *Aerococcus* PDS

	Public Health Agency of Canada	Agence de la santé publique du Canada
Home > Laboratory Biosafety and Biosecurity > Pathogen		
<b>AEROCOCCUS SPP.</b>		
<b>PATHOGEN SAFETY DATA SHEET -</b>		
<b>SECTION I - INFECTIOUS AGENT</b>		
<b>NAME:</b> <i>Aerococcus</i> spp.		
<b>SYNONYM OR CROSS REFERENCE:</b> <i>Aerococcus chr viridans</i> (1) <i>Aerococcus urinaeequi</i> (previously <i>Pedio</i>		
<b>CHARACTERISTICS:</b> Gram-positive coccoid bacteriu stains; some pairs and singles may also be found (1), non-motile, non-spore-forming, and catalase negative are absent (1), (5).		
<b>SECTION II - HAZARD IDENTIFICATION</b>		
<b>PATHOGENICITY/TOXICITY:</b> <i>A. urinae</i> and <i>A. sang</i> immunocompromised patients, and elderly men with suffering from urogenic septicemia, balaclitic soft ti		

Source: Public Health Agency of Canada



## **11.0 Occupational Medicine for Biomedical Research Program**

University of Southern California (USC)'s Occupational Medicine for Biomedical Research Program is designed to protect employees who are engaged in biomedical research by providing up-to-date prophylactic measures that include immunizations and tests as well as rapid response to accidents and illnesses that may be work-related.

This program has been developed by the Office of Environmental Health & Safety (EH&S) using information from experts including USC physicians and research faculty, the Centers for Disease Control and Prevention (CDC), the United States Department of Agriculture (USDA) and cohort institutions to develop best practices that provide state-of-the-art healthcare services.

In order to make the Occupational Medicine Program's services accessible and convenient, we share a relationship with the Keck Healthcare Centers, the Student Healthcare Centers, as well as other nearby healthcare providers.

USC is committed to maintaining a safe and healthy work environment for each employee and ensures full compliance with all applicable occupational safety and health regulations. The Biosafety Program in concert with the USC Institutional Biosafety Committee (IBC) performs a risk assessment of each research project involving biological hazards or chemical hazards common in biomedical research such as chemotherapy agents, carcinogens, and toxins of biological origin. This risk assessment determines which occupational medicine component(s) will be recommended or required.

Components of the Occupational Medicine Program are required by regulatory, advisory, and accreditation bodies in order to conduct biomedical research, including immunizations, vaccines, tests, and other procedures related to research hazards. They must be offered to employees, but are not mandatory and may be declined. In some cases, prophylactic measures are required in order for an employee to safely participate in a particular research. In that case, the employee may choose not to undertake the research project if he or she is not willing to comply with the medical recommendation.

# Animal Exposure Program

## Overview

There are a number of hazards associated with laboratory animals. These include allergens, bites and scratches, zoonoses, and chemicals and infectious microorganisms with which animals may be treated. There are other hazards associated with the animal facility itself such as cleaning compounds, and the physical hazards associated with autoclaves, cage washers, and animal housing racks. Some of these hazards may be mitigated through participation in the [Animal Exposure Program](#).

## Risks

Some studies have shown that up to 30% of those who work with laboratory animals will develop allergies within the first three years of work. Even if you don't touch or handle laboratory animals, some individuals may experience symptoms and some may develop allergies to animals (see Figure 11.1 Animal Allergies Fact Sheet). Animals generate dander from their skin, fur, and saliva and this can be a source of allergens in the air. Some people have allergies to animal dander when they breathe it in or they come in contact with it on their skin. The excrement from animals (urine and feces) also contains proteins that can cause an allergic reaction in some people.

Allergy symptoms can consist of the following: itchy eyes, nasal stuffiness, sneezing, coughing, wheezing, a tight chest, and rashes on the skin. These symptoms can range from mild to severe depending upon how allergic someone is. There are some people who may have a lot of exposure to animal allergens but never become allergic.

## Mitigation

The Animal Exposure Program provides each individual who experiences animal exposure or potential exposure an opportunity to participate in a risk assessment regarding their animal work. The completed risk assessments are reviewed by a physician who determines whether further follow up is required.

After review by a physician, EH&S's Biosafety Program is informed about the results of the evaluation. Only non-confidential information is communicated. Biosafety then contacts those who require follow-up and offers them an opportunity to speak with a USC Keck Internal Medicine physician, free-of-charge.

Figure 11.1. Animal Allergies Fact Sheet

### FactSheet

### Animal Allergies

Most employees working in animal facilities find it similar to any other work environment. However, some employees may experience allergy symptoms that can be directly related to their work with or proximity to animals. Allergies can develop and the symptoms expressed even when there is no direct contact with animals.

**WHY DO ANIMALS CAUSE ALLERGIES?**  
Proteins in animal dander and excrement (urine and feces) may cause allergic reactions. Proteins that cause allergies are called "allergens" and may exist as airborne particles in animal facilities.

**CAN ANYONE BECOME ALLERGIC TO LABORATORY ANIMALS?**  
Individuals that have existing allergies, a family history or susceptibility to allergens, or suppressed immune systems may be at a higher risk for developing allergies. Others may develop allergies over time that increase in intensity as exposure increases; some may never experience allergies.

**WHAT ARE THE SYMPTOMS OF ANIMAL ALLERGIES?**  
Allergy symptoms consist of the following: itchy eyes, nasal stuffiness, sneezing, coughing, wheezing, difficulty breathing, a tight chest, and rashes on the skin. These symptoms can range from mild to severe.

**HOW DO I PROTECT MYSELF FROM ANIMAL ALLERGIES?**  
Three levels of control are employed to prevent workers from contact with animal allergens: engineering controls, workplace practices or administrative controls, and personal protective equipment (PPE).

**Engineering controls.** For USC animal facilities, these include: biosafety cabinets, animal transfer stations, cage dumping stations, and individually ventilated caging systems. This HEPA-filtered containment equipment is maintained and calibrated to industry standards.

Air quality monitoring of high risk areas is provided by EH&S. Personal and area monitoring may be performed to determine the effectiveness of controls.

**WHAT I NEED TO DO**

- Know the signs and how to report in case of a suspected allergic reaction.
- Use engineering and administrative controls as well as appropriate PPE to reduce exposure to animal allergens.
- Complete an [ARA form](#) (see [Administrative controls](#) below).
- Wash hands after removal of protective gloves.

**Administrative controls.** These include the [Animal Exposure Program](#) and [Occupational Medicine for Biomedical Research](#). An [Animal Exposure Risk Assessment form \(ARA\)](#) that includes animal hazards, medical history, and factors that may influence allergy development is completed by the employee. An Occupational Medicine physician then reviews the completed ARA form. Based on the assessment, the following may be recommended: use of an N95 respirator, a follow-up evaluation with a physician, or both (refer to the [ARA Fact Sheet](#) for details).

**PPE.** The Department of Animal Resources (DAR) has a minimum PPE requirement for entering animal facilities that includes:

- Pants or other leg covering
- Closed-toe/closed heel shoes worn with socks
- Disposable, closed front gown
- Gloves
- Shoe covers
- Face mask
- Hair bonnet

Clothing that does not meet or exceed DAR requirements is not allowed in animal facilities.

**REFERENCES**

- [Occupational Health and Safety in the Care and Use of Research Animals, 3RD EDITION](#)
- [Guide for the Care and Use of Laboratory Animals \(NIH, 2011\)](#)
- [NIOSH: Preventing Asthma in Animal Facilities](#)

After being authorized by Biosafety, the individual makes an appointment with the Keck physician through the Internal Medicine Patient Liaison Office instead of the regular appointment scheduling process to facilitate the process.

The Department of Animal Resources provides a brief training and assurance form for those who enter animal facilities but have no contact with animals.

## Resources

The following resources are available to help you participate in the program and learn more:

- [Animal Exposure Occupational Medicine Program](#)
- [Animal Allergen Prevention Program](#)
- [Animal Exposure Risk Assessment \(ARA\)](#) - Required
- [Animal Exposure Risk Assessment Instructions](#)
- [Animal Allergies Fact Sheet](#)
- [Animal Exposure Program SOP](#)
- [Non-Contact Animal Exposure Acknowledgment Form](#)
- Keck Internal Medicine Form

## Infectious Agent and Toxins Program

### Overview

Those involved in biomedical research often use infectious disease agents to study the internal mechanisms of the agents involved in human disease. The USC IBC and Biosafety Program rely on our local infectious disease physicians, public health, and CDC and NIH Guidelines to provide the most current recommendations for the immunizations, tests, and other healthcare activities to prevent exposure to the infectious agents used in research.

### Immunizations

The Occupational Medicine Program currently offers immunization against a number of agents used in research. In some cases, titers following vaccination are recommended by the CDC Advisory Committee on Immunization Practices to ensure immunity and those are offered also.

In addition to immunizations and titers, various tests are also recommended for work with specific agents. Tests are used to identify an inadvertent exposure to an infectious disease agent. For example, those who work with *Mycobacterium tuberculosis* (Mtb) are offered either the standard skin test known as a PPD or Interferon-gamma release assays (IGRAs), relatively new diagnostic tools for latent tuberculosis infection (LTBI), can differentiate between those who have been vaccinated with BCG and those who have had Mtb exposure.

Table 11.1 provides an outline of immunizations and titers offered and Table 11.2 is an outline of various tests provided by the Office of EH&S.

As research progresses to additional agents, additional immunizations and tests may be added.

**Table 11.1. Immunizations and Tests Offered Upon Risk Assessment Completion**

Agent	Immunization	Titer
<i>Clostridium botulinum</i>	IND	
<i>Clostridium tetani</i>	Tdap or Td	
Diphtheria toxin	Tdap or Td	
Hepatitis A virus	depends	
Hepatitis B virus	HBV	Yes
Human papillomavirus	HPV	
Influenza virus	seasonal	
Measles virus	MMR	
Mumps virus	MMR	
Pertussis toxin	Tdap	
Polio virus	IPV	
Rabies virus	Pre-exposure and post-exposure	Yes
Rubella virus	MMR	
Vaccinia virus	ACAM 2000	
Yellow Fever virus	YFV	

**Table 11.2. Tests available**

Agents	Tests
Mycobacterium tuberculosis	Mtb PPD or IGRA
Hepatitis B Virus	HBV
Hepatitis C Virus	HCV
Human Immunodeficiency Virus	HIV

Note that risk group 2, risk group 3, and select agents and toxins are covered as appropriate.

## Resources

The following resources are available to help you participate in the program and learn more:

- [Exposure Control Plans](#)
- [Immunizations Form](#)
- [HBV Acceptance/Declination Form](#)

## Specific Chemicals Program

### Overview

There are a number of hazardous chemicals commonly used in biomedical research for which a biological use application (BUA) is required. These include carcinogens, chemotherapy agents, toxins of biological origin, and other chemicals that are deemed hazardous by the IBC, biosafety, or another authority. Biosafety performs a risk assessment and requires an exposure control plan or SOP for these agents. When there is a medical intervention that can be applied prior to exposure or a process for medical care upon exposure that can be developed ahead of time, the Occupational Medicine Program provides access to it for employees.

### Mitigation

Not all chemicals used in biomedical research have a prophylactic measure available. All chemical exposures, even small ones, must be reported to one's supervisor. The supervisor is responsible to complete a manager's incident report and to ensure that an injured employee seeks medical attention promptly.

**Table 11.3. BUA-Required Chemical Pre- and Post-Exposure Processes**

Chemical	Research Role	Pre-Exposure Process	Post-Exposure Process
MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine)	A neurotoxin that causes Parkinson's-like symptoms	<ul style="list-style-type: none"> <li>ECP, SOP, SDS</li> <li>Animal bedding SOP</li> <li>Physical and health history</li> <li>Prescription for eldepryl/selegiline</li> <li>Work with this agent requires approval by physician</li> </ul>	<ul style="list-style-type: none"> <li>Follow ECP</li> <li>Seek medical attention immediately</li> <li>Medication under a physician's care</li> </ul>
ENU N-ethyl-N-nitrosourea	Potent mutagen	<ul style="list-style-type: none"> <li>ECP, SOP, SDS</li> <li>Animal bedding SOP</li> </ul>	<ul style="list-style-type: none"> <li>Follow ECP</li> <li>Seek medical attention immediately</li> </ul>
<ul style="list-style-type: none"> <li>Pertussis toxin</li> <li>Diphtheria toxin</li> <li>Tetanus toxin</li> </ul>	<ul style="list-style-type: none"> <li>Used in the development of animal models for human disease</li> <li>Other uses in biomedical research</li> </ul>	<ul style="list-style-type: none"> <li>ECP, SOP, SDS</li> <li>Animal bedding SOP</li> <li>Vaccine</li> </ul>	<ul style="list-style-type: none"> <li>Follow ECP</li> <li>Seek medical attention</li> <li>Antitoxin may be available</li> </ul>
Other toxins of biological origin, e.g., Tetrodotoxin Conotoxin, SEB, others	Various uses in biomedical research	<ul style="list-style-type: none"> <li>ECP, SOP, SDS</li> <li>Animal bedding SOP</li> </ul>	<ul style="list-style-type: none"> <li>Follow ECP</li> <li>Seek medical attention</li> </ul>
Carcinogens (Prop 65 list)	Various uses in biomedical research	<ul style="list-style-type: none"> <li>ECP, SOP, SDS</li> <li>Animal bedding SOP</li> </ul>	<ul style="list-style-type: none"> <li>Follow ECP</li> <li>Seek medical attention</li> </ul>
Chemotherapy agents	Various uses in biomedical research	<ul style="list-style-type: none"> <li>ECP, SOP, SDS</li> <li>Animal bedding SOP</li> </ul>	<ul style="list-style-type: none"> <li>Follow ECP</li> <li>Seek medical attention</li> </ul>
Streptozotocin	Induces diabetes in several animals through destruction of B-cells	<ul style="list-style-type: none"> <li>ECP, SOP, SDS</li> <li>Animal bedding SOP</li> </ul>	<ul style="list-style-type: none"> <li>Follow ECP</li> <li>Seek medical attention</li> </ul>

## Resources

The following resources are available to help you participate in the program and learn more:

- Exposure Control Plans
- MPTP Plan

# Macaque Program

## Overview

Individuals who work with non-human primates are susceptible to zoonotic diseases. They are offered an annual tuberculosis screening that may be required depending upon the job duties.

## Risks

Macaques (see Figure 11.1) are considered to be potential carriers of Macacine herpesvirus 1, formerly known as Herpes B virus, a virus that macaques may carry in a latent form that can be devastating to humans.

All macaques are considered potential carriers, even if they come from a "tested" or "Herpes B virus-free" colony.

**Figure 11.1. Barbary Macaque**



Source: Wikipedia

## Mitigation

Anyone who works with or around macaques must undergo training regarding the DAR document *Procedures for Bite, Scratch, or Mucous Membrane Exposure to Macaque Nonhuman Primates* and be aware that samples must be taken from an injured individual as well as the initiating macaque and sent to the National B Virus Resource Center in cases of accidental exposure. Contact DAR for animal training and the Biosafety Program [biosafety@usc.edu](mailto:biosafety@usc.edu) for information regarding safely handling macaques and their materials as well as information regarding exposure to Macacine herpesvirus (Herpes B).

## Resources

Contact EH&S Biosafety Program for a copy of the most current macaque exposure procedures document.

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## 12.0 Bloodborne Pathogen Program

### Bloodborne Pathogens

[Cal-OSHA](#) defines bloodborne pathogens (BBPs) as infectious agents present in human blood that can cause disease in humans. BBPs exist as viruses, bacteria, or parasites. Healthcare employees, researchers, housekeeping personnel, and other workers who handle human tissues or cells are at risk for exposure to BBPs. One mode of potential BBP transmission among researchers is through needlesticks and other sharps-related injuries.

The OSHA and closely-related Cal-OSHA Standard were established to help reduce or eliminate workplace hazards that consequently lead to BBP exposures. The standards cover all pathogens that are transmitted through exposure to human or non-human primate blood. USC's Bloodborne Pathogen Program provides a description of the following components:

- Universal Precautions
- Definition of Bloodborne Pathogens
- Training
- Exposure Control Plans
- Procedural techniques

Universal Precautions is a concept that all human and non-human primate blood, cells, cell lines, tissues, organs, body parts, or other potentially infectious materials (OPIM) are regarded as infectious.

The top three viruses covered in the BBP standard are the following:

- Hepatitis B virus (HBV)
- Hepatitis C virus (HCV)
- Human immunodeficiency virus (HIV)

## Hepatitis B Virus (HBV)

Hepatitis B virus (see Figures 12.1 and 12.2) is considered the most common and very serious BBP disease. According to the CDC, it is estimated that more than one-third of the population has been infected with HBV of which 5% are chronic carriers. 25% of those carriers will develop serious liver diseases. There is an approved vaccine for HBV.

*Transmission in the General Population.* HBV infections can occur through blood transfusions, vaccinations, tattooing, or body piercing with inadequately sterilized instruments. Another common mode of transmission is through unprotected sex.

Those who are infected with HBV are also at risk of infection with hepatitis D virus (HDV), a defective RNA virus that requires the presence of HBV for replication. The co-infection of HDV typically exacerbates the symptoms caused by HBV.

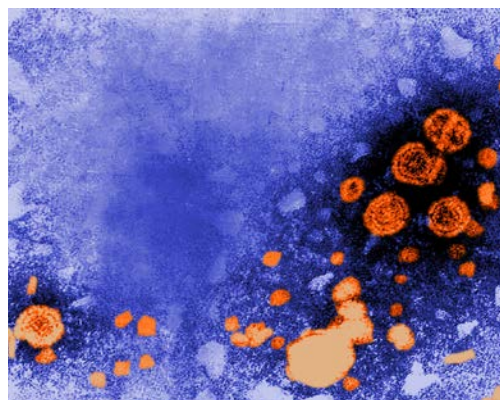
*Laboratory-Acquired Infections.* Hepatitis B laboratory-acquired infections are most frequently reported. The risk of contracting HBV is four-times greater for health care workers or laboratory personnel than that of the general public according to the CDC.

Human blood contains the highest HBV titers (number of viral particles) of all body fluids and is the most important vehicle of transmission in the healthcare industry. Avoiding occupational blood exposure reduces transmission of HBV.

*Prophylaxis, Treatments, and Vaccines.* An HBV immunization is provided by USC to any employee who uses human or non-human primate materials. In some instances, HBV titers are provided to ensure that employees are still protected.

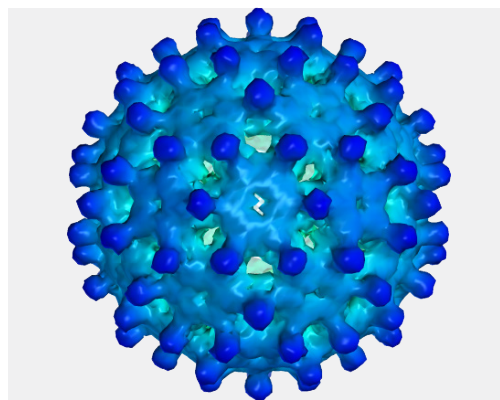
Though HBV immunization may be declined by the employee, he or she may acquire the immunization at a later time. Note that the Biosafety Program uses the HBV acceptance declination form to document that employees were offered the vaccine.

**Figure 12.1. HBV pathogen**



Source: CDC

**Figure 12.2. HBV pathogen - 3D model**



Source: NIH

## Hepatitis C Virus (HCV)

Hepatitis C virus (see Figures 12.3 and 12.4) causes both acute and chronic infections of the liver. Of those infected with HCV, 70%-85% will develop a chronic liver disease that for many will lead to liver cirrhosis or liver cancer according to the CDC.

Although an estimated 170 million people are thought to have HCV infections, the prevalence of HCV infections is < 2% in developed countries. Many infected individuals in developed countries are unaware that they have been infected.

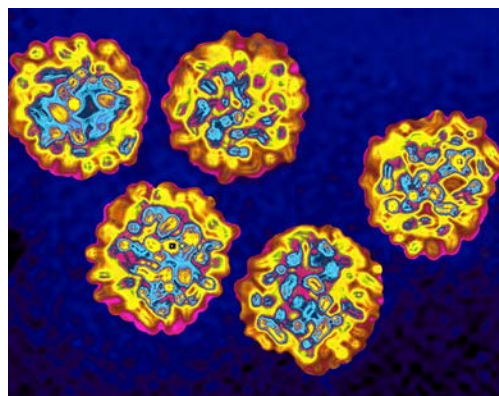
There is no vaccination for HCV, however, antiviral drugs can help cure acute HCV infections.

*Transmission in the General Population.* Hepatitis C viral infections occur most often through use of improperly sterilized needles or healthcare equipment, liver transfusions, or contact with someone already infected with HCV.

*Laboratory-Acquired Infections.* Hepatitis C virus can also be associated with laboratory infections. Needlestick injuries are the most common route of exposure in the laboratory that result in HCV infections.

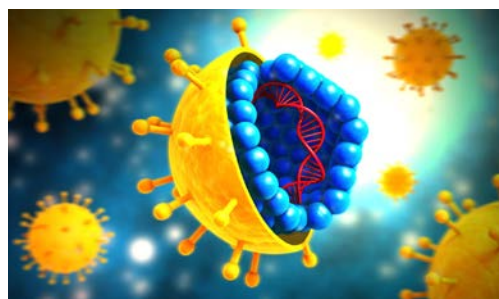
Proper use of safety-engineered needles and other controls can reduce the exposure events in the laboratory.

**Figure 12.3. HCV pathogen**



Source: NPR

**Figure 12.4. HCV pathogen - 3D model**



Source: CDC

## Human Immunodeficiency Virus (HIV)

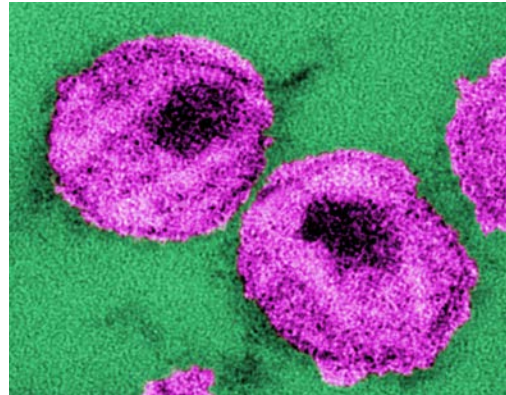
Human immunodeficiency virus (see Figures 12.5 and 12.6) continues to be a global health issue. The CDC has estimated that 54% of people with HIV know their status. HIV leads to Acquired Immunodeficiency Syndrome (AIDS) which can ultimately result in death. There is no cure for HIV nor are there any approved vaccines.

Retroviral drug therapies are the current therapy for HIV-infected people and have proven effective in delaying the onset of AIDS.

*Transmission in the General Population.* Transmission of HIV occurs through sex or contact with infected blood, semen, cervical, or vaginal fluids. Sexual contact is the most common mode of transmission worldwide. Other modes of transmission include blood transfusions (**NOTE:** The US has screened all blood donations for HIV since 1985), needles or syringes contaminated with HIV, or mother-to-infant transmission.

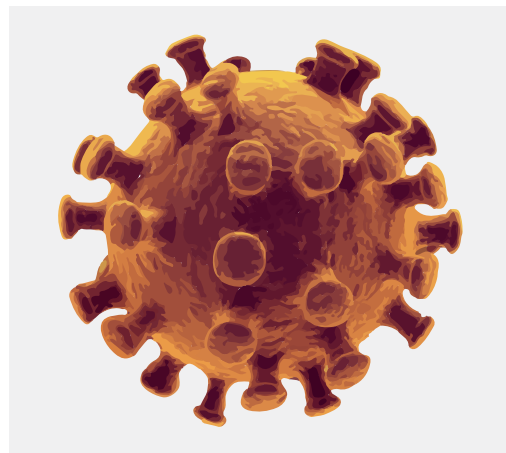
*Laboratory-Acquired Infections.* Although there were many reported laboratory-acquired infections with HIV, the overall percentage is relatively low. It is estimated that the chance of obtaining HIV through an occupational exposure is less than 0.1%, according to the CDC.

**Figure 12.5. HIV pathogen**



Source: CDC

**Figure 12.6. HIV pathogen - 3D model**



## BBP Training Requirement

All USC personnel who may have potential exposure to human blood, tissue, or cells are required to take the bloodborne pathogen training (BBP) through the EH&S Biosafety Program. Training is directed to biomedical occupations (e.g., research, dentistry, and medicine) and the type of tasks associated with them. Initial training consists of a one-hour face-to-face training session that covers bloodborne pathogens and diseases; methods used to control occupational exposure; hepatitis B vaccine; medical evaluation; and post-exposure follow-up procedures.

Subsequent refresher training is required annually per the Cal-OSHA BBP Standard. BBP and other EH&S training are available and accessed through TrojanLearn. Records of initial training and annual refresher are managed by EH&S. PIs or designees that provide annual refresher training will submit sign-in sheets to EH&S at [safetytraining@usc.edu](mailto:safetytraining@usc.edu).

## Exposure Control Plan (ECP)

According to OSHA and Cal-OSHA Bloodborne Pathogen Standards, employers must establish a written exposure control plan (ECP). The written document includes: (a) job classifications and tasks that may result in potential exposures to BBP, (b) best practices that can help minimize exposures, and (c) emergency response protocol when there is a suspected exposure.

Important concepts in exposure control are Universal Precautions and control measures. All human and non-human primate blood, cells, cell lines, tissues, organs, body parts, or other potentially infectious materials (OPIM) are regarded as infectious under the Universal Precautions paradigm.

Control measures are the engineering controls, administrative controls, and personal protective equipment (PPE) employed to effectively safeguard against exposure (see [Section 7 Biosafety Practices: Personal Protective Equipment](#)).

Engineering controls reduce the risk of exposure to BBP. Some examples are sharps containers, needle protection devices (see Figure 12.7), self-sheathing needles, and biosafety cabinets (see Figure 12.8).

**Figure 12.7. Needle protection devices**



Source: BD

**Figure 12.8. Biosafety Cabinet**



Source: ESCO

Administrative controls are policies, standard operating procedures (SOP), and best practices for the handling, containment, and disposal of infectious materials. PPE is the clothing and safety equipment used to protect the worker from biohazardous materials in the workplace. Appropriate PPE must be provided by the employer to the employee.

This includes gloves, lab coats, disposable gowns, safety eye-wear, and other equipment as needed.

Prophylactic measures for exposure to bloodborne pathogens include the hepatitis B vaccine. USC employees that risk exposure to HBV in their workplace may receive the vaccine free-of-charge. In addition, USC offers post-exposure medical follow-up free- of-charge after a work-related exposure incident. An exposure incident is a specific eye, mouth, other mucous membrane, non- intact skin, or parenteral contact with human or non-human primate blood or other materials as described above. Depending on the circumstances of the exposure incident, the employee may be offered any of the options below:

- Post-exposure prophylaxis for infectious agent
- Counseling
- Evaluation of reported disease symptoms
- Any other appropriate medical response

The medical professional will provide a limited written opinion to the employer and all diagnoses must remain confidential.

Labeling and hazard communication is a necessary component of the ECP. Those who work with bloodborne pathogens must communicate the risks with the appropriate use of signs and labels.

Laboratories that contain infectious agents must have signs on the entry doors to identify the agents, biosafety level for the lab, and proper PPE for the lab. Additionally, the following must be labeled (see Figure 12.9):

- Containers for storage or transport of contaminated materials
- Waste receptacles including sharps containers that contain contaminated materials
- Equipment used with infectious agents

**Figure 12.9. Biohazard label**



Hepatitis B vaccination records are managed by the Occupational Medicine Program for Biomedical Research. Sharps injuries records are kept by the EH&S Occupational Health Program per the Cal-OSHA BBP Standard.

#### **References**

[EH&S BBP Exposure Control Plan](#)  
[OSHA Bloodborne Pathogens \(BBP\) Standard - 29 CFR 1910.1030](#)  
[Cal-OSHA Bloodborne Pathogens Standard \(8CCR Sec. 5193\)](#)



## 13.0 Emergency Response/ Injury & Incident Reporting

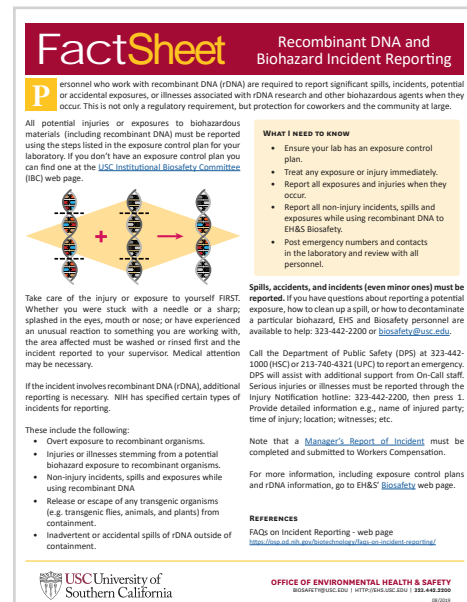
### Minor Spill

A minor spill is considered one that contaminates small areas or equipment, but DOES NOT result in external or internal contamination of personnel or serious delay in work procedures. See the [Biohazardous Spill Clean-Up Guide Sheet](#) for information.

#### Minor Spill Clean-up

1. Notify all personnel that a spill has occurred.
2. Wear PPE (including two layers of disposable gloves).
3. Cover the spill with absorbent paper and/or pads (dampen paper or pad if solids are spilled).
4. Apply bleach solution (1:10) slowly beginning at the outer periphery of the spill and moving concentrically towards the center. Allow bleach to sit for at least 10 minutes.
5. Carefully collect all absorbent paper and/or pads and deposit directly in the biohazard red bag/container. Dispose of all other contaminated materials similarly (such as disposable gloves).
6. Document the incident in email to [biosafety@usc.edu](mailto:biosafety@usc.edu). This is especially important for incidents involving recombinant DNA (rDNA) since it is monitored by NIH. Consult the [Reporting rDNA Incidents Fact Sheet](#) (see Figure 13.1) for information regarding spills of recombinant material.

Figure 13.1. Reporting rDNA Incidents Fact Sheet



<http://tiny.cc/usc-rDNA-biohz-rpt>

## Major Spill

A major spill may result in contamination of large surface areas, internal or external contamination of personnel, and/or serious delay in work procedures.

### Major Spill Clean-Up

DO NOT attempt to clean up a major spill if you do not have the proper training, resources, or confidence.

1. Notify all personnel that a spill has occurred.
2. Evacuate personnel from the area and deny entry.
3. Notify Biosafety and laboratory supervisor immediately.
4. Wear PPE (including two layers of disposable gloves).
5. Cover the spill with absorbent paper or pads, but DO NOT attempt to clean it up. Confine the movements of all potentially contaminated personnel to prevent the spread of contamination.
6. Decontaminate personnel by removing contaminated clothing and flushing contaminated skin with lukewarm water followed by washing with a mild soap.
7. Notify DPS (213) 740-4321 of the incident.

## Biohazardous Material Exposure

### Eye/Skin Contact

1. For small areas of incidental contact (except eyes), wash with mild soap and lukewarm water.
2. For large areas of contact and/or eye exposure, immediately go to the emergency shower/eye wash facility. Activate the shower and remove all contaminated clothing.
3. Flush affected body area with water for at least 15 minutes.
4. If the eyes are contaminated, forcibly hold them open and flush for at least fifteen (15) minutes.
5. Resume flushing area with water if pain continues.
6. Notify [biosafety@usc.edu](mailto:biosafety@usc.edu).

## Serious Injury or Illness Reporting

For a work-related injury or illness that requires emergency response, follow the procedures on the [Emergency Notification and Incident Reporting](#) web page, and post the [1-2-3 Serious Injury Reporting](#) flier (See Figure 13.2) in your unit's common areas to help your team become familiar with the process. Contact [EHS@usc.edu](mailto:EHS@usc.edu) for printed copies of the poster. **NOTE:** Work-related injuries and illnesses may be treated at [USC-Approved Medical Facilities](#).

Managers and HR Partners must immediately report serious occupational injuries or illnesses to EH&S so they may notify Cal-OSHA within the required eight (8) hours.

Employers who fail to report serious occupational injury or illness within eight hours are subject to a \$5,000 penalty.

**Figure 13.2. 1-2-3 Emergency Notification**



Source: EH&S - <http://tiny.cc/usc-123>

## Non-Serious Injury or Illness Reporting

Even if an injury or illness does not meet the requirements for Cal-OSHA reporting, it is important that the affected employee receives proper care. Review the [Injury/Illness: Seeking Medical Treatment Guide Sheet](#) (See Figure 13.3) for the full process.

**Figure 13.3. Injury/Illness: Seeking Medical Treatment Guide Sheet**

**GuideSheet Injury/Illness: Seeking Medical Treatment**

**EMERGENCIES**  
In the event of an emergency such as a spill, incident involving hazardous materials, an injury, or illness, call DPS at (213) 740-4321.

**FACULTY, STAFF, VOLUNTEER, OR STUDENT WORKERS WHO BECOME INJURED/ILL WHILE PERFORMING WORK FOR USC**

1. Notify your supervisor, principal investigator (PI), or instructor.
2. Seek medical care at a USC-authorized medical facility (see right-hand column of this guide sheet for a complete list). **NOTE:** bring a completed [Workers' Compensation Claim Form \(DWC-4\)](#).
3. Call Broadgate at (800) 495-2333. Broadgate is USC's worker's compensation administrator.
4. Make sure your supervisor completes a [Manager's Report of Incident Form](#).

**USC STUDENTS WHO BECOME INJURED/ILL SHOULD:**

1. Contact Engemann Student Health Center (USPC) at (213) 740-8955, or Eric Cohen Student Health Center (USCS) at (323) 442-3433 during regular business hours.
2. After business hours, contact Engemann Student Health Center and follow recorded instructions to connect with an on-call physician or nurse.

**NON-USC PARTY (I.E., VISITOR OR PARENT) INJURED ON USC PROPERTY**

1. Contact USC Risk Management at (213) 740-4209.

**USC-AUTHORIZED MEDICAL FACILITIES**

UNIVERSITY PARK CAMPUS	HEALTH SCIENCES CAMPUS
<b>Engemann Student Health Center (USPC)</b> 3551 West 44 <sup>th</sup> Street, 2 <sup>nd</sup> Floor Los Angeles, CA 90089 (213) 740-8955 (800) 495-2333	<b>USC Internal Medicine (IMC)</b> 1120 San Pablo Street, Suite 1000 Los Angeles, CA 90088 (323) 442-3200
<b>Kaiser On The Job</b> 1525 North Edgemont Street, 2 <sup>nd</sup> Floor Los Angeles, CA 90027 (323) 783-6821	<b>Eric Cohen Student Health Center</b> 1525 North Edgemont Street, 2 <sup>nd</sup> Floor Los Angeles, CA 90027 (323) 442-3433 (704) 370-0675 (DNR)

**AFTER BUSINESS HOURS AND WEEKENDS**

<b>Good Samaritan Hospital</b> Emergency Rooms 1228 Wilshire Blvd Los Angeles, CA 90017 (213) 977-2123	<b>White Memorial Medical Center</b> 1321 Wilshire Blvd Los Angeles, CA 90019 (323) 220-9875
<b>Providence Work Health Services (open 24/7)</b> 132 Broadway Ave Los Angeles, CA 90012 (213) 977-9300	<b>Kaiser On The Job</b> 1525 North Edgemont Street, 2 <sup>nd</sup> Floor Los Angeles, CA 90027 (323) 783-6821
	<b>USC Verdugo Hills Hospital</b> 1525 Verdugo Blvd Glendale, CA 91208 (818) 950-3900

**RESOURCES**

USC Injury and Illness Prevention Plan (IIPP) - <http://tiny.cc/usc-iipp>  
USC Incident Health - <http://tiny.cc/usc-incident-health>  
USC Workers' Compensation - <http://tiny.cc/usc-workers-comp>  
USC 1-2-3 Serious Injury Reporting - <http://tiny.cc/usc-123>  
Manager's Report of Incident Form - <http://tiny.cc/usc-report-incident>

**USC Environmental Health & Safety**

University Park Campus 3424 South Grand Ave. CAL 120 Los Angeles, CA 90089-2815	Health Sciences Campus 2001 N. Soto St. 58A, 529 Los Angeles, CA 90032	323.442.2386 injuryreport@usc.edu http://ehs.usc.edu
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6/2008

Source: EH&S - <http://tiny.cc/fs-usc-med-treatment>

## Near Misses

A near miss is an unanticipated event that did not result in harm or injury, but had the potential to do so. Examples would be:

- Biohazardous contamination of a large area or areas beyond a laboratory's resources to contain it
- Sudden animal recoil during injection procedure that causes the syringe needle to dislodge and drop without contact
- An open flame that is inadvertently left unattended
- Loss of: radioactive material, biohazardous agents, controlled substances, and precursor chemicals from the lab (including associated waste)

Notify Biosafety ([biosafety@usc.edu](mailto:biosafety@usc.edu) or 323-442-2200) immediately when any of these events occur. The BSO will maintain a database of accidents and near misses for educational purposes to increase user awareness of potentially hazardous situations.

## Accident Investigation

Upon notification of an accident, the Biosafety Team will conduct an accident investigation which includes the following:

- Interviews with injured workers and witnesses
- Examination of the workplace for factors associated with the accident/exposure/near miss
- Determination of possible cause(s) of the accident/exposure/near miss
- Corrective action(s) to prevent the accident/exposure/near miss from recurring
- Documentation of the findings and corrective actions taken



## Appendix A HHS and USDA Select Agents and Toxins

### HHS SELECT AGENTS AND TOXINS

- Abirn
- *Bacillus cereus* Biovar *anthracis*\*
- Botulinum neurotoxins\*
- Botulinum neurotoxin producing species of *Clostridium*\*
- Conotoxins (Short, paralytic alpha conotoxins containing the following amino acid sequence X1CCX2PACGX3X4X5X6CX7)<sup>1</sup>
- *Coxiella burnetii*
- Crimean-Congo haemorrhagic fever virus
- Diacetoxyscirpenol
- Eastern Equine Encephalitis virus<sup>3</sup>
- Ebola virus\*
- *Francisella tularensis*\*
- Lassa fever virus
- Lujo virus
- Marburg virus\*
- Monkeypox virus<sup>3</sup>
- Reconstructed replication competent forms of the
- 1918 pandemic influenza virus containing any portion of the coding regions of all eight gene segments (Reconstructed 1918 Influenza virus)
- Ricin
- *Rickettsia prowazekii*
- SARS-associated coronavirus (SARS-CoV)
- Saxitoxin
- South American Haemorrhagic Fever viruses: Chapare; Guanarito; and Junin
- Machupo and Sabia
- Staphylococcal enterotoxins A,B,C,D,E subtypes
- T-2 toxin
- Tetrodotoxin
- Tick-borne encephalitis complex (flavi) viruses:
- Far Eastern subtype and Siberian subtype
- Kyasanur Forest disease virus
- Omsk hemorrhagic fever virus
- Variola major virus (Smallpox virus)\*
- Variola minor virus (Alastrim)\*
- *Yersinia pestis*\*

### OVERLAP SELECT AGENTS AND TOXINS

- *Bacillus anthracis*\*
- *Bacillus anthracis* Pasteur strain
- *Brucella abortus*
- *Brucella melitensis*
- *Brucella suis*
- *Burkholderia mallei*\*
- *Burkholderia pseudomallei*\*
- Hendra virus
- Nipah virus
- Rift Valley fever virus
- Venezuelan equine encephalitis virus<sup>3</sup>

### USDA SELECT AGENTS AND TOXINS

- African horse sickness virus
- African swine fever virus
- Avian influenza virus<sup>3</sup>
- Classical swine fever virus
- Foot-and-mouth disease virus\*
- Goat pox virus
- Lumpy skin disease virus
- *Mycoplasma capricolum*<sup>3</sup>
- *Mycoplasma mycoides*<sup>3</sup>
- Newcastle disease virus<sup>2,3</sup>
- Peste des petits ruminants virus
- Rinderpest virus\*
- Sheep pox virus
- Swine vesicular disease virus

### USDA PLANT PROTECTION AND QUARANTINE (PPQ) SELECT AGENTS AND TOXINS

- *Coniothyrium glycines*
- *Peronosclerospora philippinensis*
- (*Peronosclerospora sacchari*)
- *Ralstonia solanacearum*
- *Rathayibacter toxicus*
- *Sclerophthora rayssiae*
- *Synchytrium endobioticum*
- *Xanthomonas oryzae*\*

## HHS and USDA Select Agents and Toxins Table Notes

\* Denotes Tier 1 Agent

- 1 C = Cysteine residues are all present as disulfides, with the 1st and 3rd Cysteine, and the 2nd and 4th Cysteine forming specific disulfide bridges; The consensus sequence includes known toxins  $\alpha$ -MI and  $\alpha$ -GI (shown above) as well as  $\alpha$ -GIA, Ac1.1a,  $\alpha$ -CnIA,  $\alpha$ -CnIB; X1 = any amino acid(s) or Des-X; X2 = Asparagine or Histidine; P = Proline; A = Alanine; G = Glycine; X3 = Arginine or Lysine; X4 = Asparagine, Histidine, Lysine, Arginine, Tyrosine, Phenylalanine or Tryptophan; X5 = Tyrosine, Phenylalanine, or Tryptophan; X6 = Serine, Threonine, Glutamate, Aspartate, Glutamine, or Asparagine; X7 = Any amino acid(s) or Des X and; "Des X" = "an amino acid does not have to be present at this position." For example if a peptide sequence were XCCHPA then the related peptide CCHPA would be designated as Des-X.
- 2 A virulent Newcastle disease virus (avian paramyxovirus serotype 1) has an intracerebral pathogenicity index in day-old chicks (*Gallus gallus*) of 0.7 or greater or has an amino acid sequence at the fusion (F) protein cleavage site that is consistent with virulent strains of Newcastle disease virus. A failure to detect a cleavage site that is consistent with virulent strains does not confirm the absence of a virulent virus.
- 3 Select agents that meet any of the following criteria are excluded from the requirements of this part: Any low pathogenic strains of avian influenza virus, South American genotype of eastern equine encephalitis virus, west African clade of Monkeypox viruses, any strain of Newcastle disease virus which does not meet the criteria for virulent Newcastle disease virus, all subspecies *Mycoplasma capricolum* except subspecies *capripneumoniae* (contagious caprine pleuropneumonia), all subspecies *Mycoplasma mycoides* except subspecies *mycoides* small colony (Mmm SC) (contagious bovine pleuropneumonia), and any subtypes of Venezuelan equine encephalitis virus except for Subtypes IAB or IC, provided that the individual or entity can verify that the agent is within the exclusion category. 9/10/13
- 4 For determining the regulatory status of nucleic acids that are capable of producing infectious forms of select agent viruses, reference guidance at <https://www.selectagents.gov/na-guidance.html>.
- 5 For determining the regulatory status of Recombinant and/or Synthetic nucleic acids that encode for the toxic form(s) of any select toxins if the nucleic acids (i) can be expressed *in vivo* or *in vitro*, (ii) are in a vector or recombinant host genome and can be expressed *in vivo* or *in vitro*; reference guidance at <https://www.selectagents.gov/na-guidance.html>.



## Appendix B Forms and Templates

### Inspections

Biosafety Inspection Process Flow Chart	<a href="#">Biosafety Inspection Process</a>
Biosafety Inspection Checklist	<a href="#">Biosafety Inspection Checklist</a>
PPE Inspection Checklist	<a href="#">PPE Inspection Checklist</a>

### Reporting

Workers' Compensation Form	<a href="#">WC form</a>
Manager's Incident Report	<a href="#">Report of Incident</a>
Volunteer Injury or Illness Report	<a href="#">Volunteer II Report</a>
Report a Safety Concern Online Form	<a href="#">Report a Safety Concern</a>

### Other

Site-Specific Training Record Form	<a href="#">Site-Specific Training Record</a>
Employee Training Record Form	<a href="#">Employee Training Record</a>

