i. Review & Signature Page

This version of this procedure manual has been reviewed for regulatory compliance and best management practices by the undersigned individuals and is hereby adopted for use and compliance by all employees at the University of Texas at San Antonio owned or operated facilities.

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I. Overview and Purpose  
This plan was prepared by the Environmental Health Safety and Risk Management (EHSRM) office after review of pertinent federal and state regulatory requirements from the Occupational Health and Safety Administration (OSHA), the Texas Department of State Health Services, the Texas Commission on Environmental Quality (TCEQ) as well as guidelines required by the Centers for Disease Control and Prevention (CDC) [http://www.cdc.gov/biosafety/publications/bmbl5/index.htm](http://www.cdc.gov/biosafety/publications/bmbl5/index.htm) and National Institutes of Health- NIH Guidelines for Research Involving Recombinant DNA Molecules [http://oba.od.nih.gov/oba/rac/Guidelines/NIH_Guidelines.htm](http://oba.od.nih.gov/oba/rac/Guidelines/NIH_Guidelines.htm). Research and education in science laboratories involve a variety of hazards. It is The University of Texas at San Antonio’s (UTSA) policy to protect and promote the health and safety of students and employees as well as the environment. This plan outlines basic good laboratory safety practices, special procedures for this institution, federal and state guidelines, and references to other information sources for work in laboratories that handle, use or store biological agents. It is not intended to be a fully comprehensive reference but rather a guidebook. There may be agents, procedures and other circumstances in each laboratory that present unique or unusual hazards not addressed here; these hazards are best addressed by the principal investigator or supervisor of the respective laboratory in consultation with EHSRM if necessary.

Faculty, staff and students who may be exposed to biological hazards in the laboratory should be informed of the nature of these hazards and how to protect themselves and others who may also be exposed. Safety in the laboratory can be achieved only with the exercise of sound judgment and proper use of facilities by informed, responsible individuals.

II. Scope  
This plan applies to all UTSA operated (leased or owned) facilities and equipment (including vehicles). It also applies to any UTSA employee, volunteer or student worker who works directly with, or is in close proximity to anyone conducting research which falls under federal and state regulations or guidelines for working with biological agents. The NIH guidelines are mandatory for all researchers at institutions receiving NIH funding, even if those researchers are not receiving NIH funding, and therefore apply to all UTSA research. All federal NIH funding can be removed from an institution for violations of the guidelines by any researcher.

III. Periodic Review  
This plan will be reviewed as needed, but at least once every 3 years for relevance and regulatory updates. The online version of this plan should be reviewed periodically for updates on the EHSRM website at: [http://www.utsa.edu/safety/](http://www.utsa.edu/safety/). Questions can be addressed to the Laboratory Safety Manager who also serves as the Institutional Biosafety Officer through EHSRM at 458-5250.

IV. Responsibilities  
A. Environmental Health Safety and Risk Management (EHSRM) will:

1. Establish the general policies and standards for the use of biological hazards at UTSA in conjunction with the Institutional Biosafety Committee (IBC), Laboratory Safety Committee, and as per directives in HOP 9.05 – Occupational Safety &
Health and 9.22 – Acquired Immune Deficiency Syndrome, Human Immunodeficiency Virus and Hepatitis B Virus (http://utsa.edu/hop/chapter9/).

2. Provide consulting services for work with biological agents.

3. Review applications and protocols for work with potentially infectious materials or hazardous biological agents and provide recommendations to the Principal Investigator (PI), Institutional Biosafety Committee (IBC), Institutional Review Board (IRB), Institutional Animal Care and Use Committee (IACUC), or University Veterinarian.

4. Develop safety plans and training programs for work with all risk groups of biological agents, bloodborne pathogens, and other potentially infectious materials in use at UTSA facilities.

5. Contract for the annual certification, maintenance and repair of biological safety cabinets to ANSI/NSF-49.

6. Maintain a biological waste disposal program.

7. Supervise decontamination and clean-up activities following spills or exposures.


9. Maintain qualified staff to act as the Responsible or Alternate Responsible Official for Select Agent Program work at UTSA and as the Institutional Biosafety Officer.

10. Investigate biological exposure incidents.

11. Evaluate laboratories periodically to ensure compliance with institutional, state and federal guidelines and regulations as they pertain to biosafety.

12. Provide the final clearance for the safe demolition, renovation or reassignment of UTSA facilities and equipment that used or contained hazardous biological agents or potentially infectious materials.

B. Principal Investigators (PI) or Laboratory Supervisors will:

1. Submit protocols for all non-exempt biological work to the IBC and await approval prior to conducting work covered by the protocol.

2. Enforce all UTSA procedures and policies regarding all risk groups of biological agents.

3. Ensure laboratory personnel have been properly trained to work safely within their laboratory to include required safety training provided by EHSRM.

4. Develop specific safety procedures or protocols for their laboratory.

5. Advise EHSRM of any significant protocol changes and prior to bringing new biologically hazardous agents onto campus.

6. Report any exposures, spills, thefts or other incidents involving biological safety to EHSRM immediately or as soon as possible.
7. Maintain a clean and sanitary workplace.
8. Report any plans to remodel or alter UTSA Facilities (HOP 8.3) to Facilities and EHSRM and gain permission before proceeding.

C. Laboratory Staff or Worker will:
   1. Observe the established guidelines, protocols and policies for biological safety.
   2. Attend all necessary or required training.
   3. Report all spills or incidents to their supervisor and to EHSRM if necessary.
   4. Report to the supervisor or EHSRM any unsafe practices or conditions in the laboratory.
   5. Properly dispose of all laboratory wastes.

D. The Institutional Biosafety Committee (IBC) will:
   1. Assist in approving general policies and procedures for the biohazards at UTSA.
   2. Review and exercise approval authority of protocols provided by principle investigators relating to the use of biohazards and recombinant DNA.
   3. Maintain the required records of the protocol review, approval and monitoring of the use and disposal of biohazards, as required by the NIH recombinant DNA guidelines.
   4. Serve as an avenue of appeal in case of dispute between EHSRM and the PI.

E. The Laboratory Safety Committee will:
   1. Assist in reviewing new safety issues involving laboratories.
   2. Review facility safety issues involving laboratories.
   3. Review continuing safety issues involving laboratories.

V. Biological Laboratory Safety Plan
   A. Biosafety

Safety is very important in biological laboratories due to the microscopic nature of many of the organisms being studied, the high concentrations involved, and the infectious nature of many organisms. History has shown that workers in laboratories working with microorganisms have become infected by the organisms they are working with for years. Such incidents continue to occur. These infections are known as laboratory acquired infections (LAI). LAI’s may follow a route of infection different from that in nature.

Biosafety is the application of combinations of laboratory practice and procedure, laboratory facilities, and safety equipment when working with potentially infectious microorganisms. Following biosafety practices provide protection for laboratory
workers (yourself), the products you are working with, co-workers within the laboratory, people outside the laboratory (including families) and the environment. Chemicals are also used in biological laboratories so chemical safety must also be observed (refer to the UTSA Chemical Safety Plan).

B. Safety guidelines

1. No mouth pipetting, use only pipetting devices.
2. No food or drink stored or consumed in the laboratory.
3. Wash hands frequently, especially after known contamination, after removing gloves and before leaving the work area or touching common use items such as computer keyboards, telephones, or door handles.
4. Remove gloves before touching common use items.
5. Syringes, Needles and Sharps
   a. Plastic ware should be substituted for glassware whenever possible.
   b. Whenever practical, laboratory supervisors should adopt improved engineering and work practice controls that reduce the risk of sharps injuries.
      1) Examples of engineering controls include: self-sheathing needles, needleless systems, and blunt tipped equipment such as scalpels and scissors.
      2) Examples of work practice controls include: not recapping needles and pointing sharps away from the user especially when carrying out an injection.
   c. Non-disposable sharps must be placed in a hard walled container for transport to a processing area for decontamination, preferably by autoclaving.
   d. Syringes should be handled with great care and only after adequate training.
      1) After filling, excess fluid and bubbles should be expelled from syringes vertically into a plastic test tube containing a sterile cotton pledget to minimize aerosolization or this procedure can be performed in a class II biosafety cabinet.
      2) Contaminated needles and syringes will be discarded in the sharps container. DO NOT RECAP CONTAMINATED NEEDLES. ONCE DISCARDED, ITEMS MUST NOT BE REMOVED FROM THE SHARPS CONTAINER.
      3) Never bend, shear, break, remove from disposable syringes or otherwise manipulate by hand needles prior to disposal.
      4) Needles used for blood drawing (phlebotomy) should also be placed in an appropriate sharps disposal container. DO NOT RECAP CONTAMINATED NEEDLES.
5) Any sharps injury must now be reported to EHSRM via the First Report of Injury Form available at the department’s website.

6. Broken Glass

Clean up broken glass as soon as possible to prevent injuries. Collect broken glass using a broom and dust pan where possible. Inside a BSC or other piece of equipment tongs or forceps can be used to collect the broken glass. NEVER use hands to pick up broken glass even if gloves are worn. Place any broken glass in a puncture-resistant container. When this container is approximately three quarters full, seal the container and dispose of it or make arrangements with Housekeeping to dispose of it.

7. Bio-aerosol formation should be avoided or minimized. Bio-aerosols are formed when the liquid-air interface is disturbed. Inhalation of bio-aerosols can lead to infections even by organisms not known to be transmitted by the aerosol route. Prevention of bio-aerosols hazards:

a. Use absorbent paper on workbench

b. Perform all bio-aerosol forming procedures inside a BSC or substitute with other procedures. Some bio-aerosol forming laboratory activities: opening centrifuge tubes, flaming loops, blowing the last drop out of pipets, splashes, vortexing, operating a flow cytometer, working with a cryostat and operating a cell sorter.

C. rDNA—Recombinant DNA

Recombinant DNA molecules are defined as either: molecules that are constructed outside living cells by joining natural or synthetic DNA segments to DNA molecules that can replicate in a living cell, or molecules that result from the replication of those described above. Work with rDNA must be approved by the Institutional Biosafety Committee unless it is exempt under the NIH guidelines (http://oba.od.nih.gov/oba/rac/Guidelines/NIH_Guidelines.htm). A listing of exempt experiments is listed below. If you have consulted the NIH guidelines and the listing below and still have questions, contact the Institutional Biosafety Officer in EHSRM. Be aware that some experiments considered exempt by NIH will require a protocol submission to the UTSA IBC per UTSA policy.

1. Exempt experiments

a. The rDNA is never going to be in an organism or virus.

b. The rDNA is solely from a single non-chromosomal or viral source.

c. The rDNA is solely from a prokaryotic host and propagated in the same host or transferred to another host by well established physiological means.

d. The rDNA is from a eukaryotic host and is propagated in the same host.

e. The rDNA is from a species that naturally exchanges DNA by known physiological processes. A list of species are available at the NIH website.
f. The rDNA is of a type which does not present a significant risk to health or the environment, as determined by the NIH Director*.

*The NIH has determined that rDNA from infectious agents of BSL-2 or above is not exempt and must receive IBC approval. Additionally, certain cloning vectors, such as Adeno or Sindbis based vectors, or amphotrophic MMLV based vectors, are some examples that are nonexempt.

2. Experiments Requiring Prior Approval

The following experiments require prior approval from the NIH, Recombinant DNA Advisory Committee (RAC), Food and Drug Administration, and/or the IBC.

a. Gene transfer experiments in humans;

b. Genes for toxins lethal for vertebrates;

c. Release of genetically engineered organisms to the environment;

d. Those using human or animal pathogens (biosafety level 2 and higher) as host-vector systems, including adenovirus vectors and murine retroviruses that infect human cells;

e. Cloning DNA from human or animal pathogens (biosafety level 2 and higher) into a non-pathogen host-vector system;

f. Cultures of more than 10 liters; and,

b. Experiments involving whole plants or animals, including transgenic organisms.

3. Experiments Requiring IBC Notice Simultaneous with Initiation

Some recombinant DNA work requires IBC review and approval, but prior approval is not required, and may be conducted at BSL-1 containment. Examples include:

a. Recombinant DNA molecules containing no more than 2/3 of the genome of any eukaryotic virus (with some restrictions) propagated and maintained in cells in tissue culture. It must be demonstrated that the cells lack helper virus for the specific families of defective viruses being used. Many, but not all, experiments involving whole plants.*

* Some plant experiments do not require prior approval. Work with recombinant DNA in plants or any work with plant pathogens must also comply with USDA and EPA regulations.

D. Working with Potentially Infectious Agents

Infectious agents are viable microorganisms, or their toxins, which cause or may cause disease in humans or animals. Examples of infectious agents include bacteria, viruses, fungi, parasites and prions. Prions are infectious proteins.
Infectious agents are classified into four risk groups (RG) according to the severity of their effects on human health. Appendix I gives examples of agents in each risk group.

1. Risk Groups:
   a. Risk Group 1 - Agents that are not associated with disease in healthy adults.
   b. Risk Group 2 – Agents that are associated with human disease which is rarely serious and for which preventive or therapeutic interventions are often available.
   c. Risk Group 3 – Agents that are associated with serious or lethal human disease for which preventive or therapeutic interventions may be available (high individual risk, low community risk)
   d. Risk Group 4 – Agents that are likely to cause serious or lethal human disease for which preventive or therapeutic interventions are not usually available (high individual risk and high community risk)

2. Biological Safety Levels: Biological Safety Level (BSL) or Biosafety Level (BL) refers to the actions, precautions, and equipment needed to protect people and the environment from biological agents being worked with in the laboratory.
   a. BSL-1 involves agents which are not known to cause disease in healthy adults. This is the level that teaching laboratories work at for the most part. Few special precautions are necessary when working at this level. However, glove usage is recommended.
   b. BSL-2 involves agents that are known to cause disease in humans. The diseases are not known to result in major illnesses in healthy adults. Some precautions are necessary when working at this level. Gloves must be worn, laboratory coats are strongly suggested. Any work that can result in the creation of bio-aerosols, should be carried out in the appropriate biological safety cabinet (BSC).
   c. BSL-3 involves agents that are known to cause disease through the aerosol route. These diseases are usually not communicable, but they do result in serious diseases. They have a high morbidity rate, but the mortality rate is not high.
   d. BSL-4 involves agents that cause serious diseases which have high mortality rates and for which there is no known cure. There are very few laboratories in the U.S. that operate at this level. UTSA does not have a laboratory of this type.

3. Requirements for facilities and practices at BSL-1.
   a. Standard Microbiological Practices
      1) Limited or restricted access to the laboratory when work is in progress.
      2) Biohazard warning signs must be in place.
3) No eating, drinking or smoking in laboratory.
4) No mouth pipetting.
5) Hands must be washed after handling viable materials and when leaving the laboratory.
6) Efforts to minimize splashes and aerosols must be made.
7) Work surfaces must be decontaminated daily and immediately after spills.
8) Wastes must be decontaminated.
9) An insect and rodent control program must be maintained.
10) Personal protective equipment will be required at the discretion of the laboratory supervisor.

b. No Special Practices Required

c. Secondary Barrier – Facility Requirements: Doors for the laboratory, hand washing sink available, work surfaces made from easily cleanable material, bench tops made from material impervious to water, sturdy furniture, and any windows are fitted with fly screens.

d. No special facilities required

4. BSL-2: Standard Microbiological Practices

a. All BSL-1 Practices Plus:

Use of Class II Biosafety Cabinets (BSC) for work with infectious agents involving aerosols and splashes, large volumes or high concentrations.

b. BSL-2: Special Practices

1) Policies and procedures must be in place for workers to gain entry.
2) Laboratory specific biosafety manual must be written by the PI or laboratory supervisor.
3) Training, including annual updates, must be provided for laboratory specific issues and SOP’s.
4) Leak-proof transport containers are necessary.
5) Immunizations against agents being worked with must be available to laboratory staff.
6) Baseline serum samples should be banked.
7) Work surfaces must be decontaminated.
8) Spills and accidents must be reported.
9) No animals or plants in this laboratory which are not part of the research.

10) Sharps precautions must be followed including, dispose of sharps in a sharps’ container, never recap, bend, break or reuse needles or syringes and never use hands to pick up broken glass when placing it in a broken glass container.

c. Secondary Barrier – Facility Requirements: adequate illumination, eyewash inside the laboratory, laboratory pressure negative to the hallway, air from the laboratory cannot be recirculated within the building, the door must be lockable to limit access when work is in progress, an autoclave must be available and the lab must be separated from public areas of the building.

5. BSL-3: Standard Microbiological Practices

   a. All BSL-1 and/or BSL-2 Practices Plus: All manipulations of infectious agents must be carried out inside a class II or III BSC.

      Personal Protective Equipment for BSL-1 and BSL-2 and respiratory protective equipment as indicated.

   b. BSL-3: Special Practices

      1) BSL-2 Special Practices Plus: When carrying out bio-aerosol forming procedures, bio-aerosol containing equipment must be used.

      2) All spills must be promptly decontaminated.

   c. Secondary Barrier – Facility Requirements: All requirements for BSL-1 and BSL-2 Plus; the laboratory should be in a separate building or in an isolated zone, double door entry into the laboratory, single pass air with 10-12 air changes per hour, room penetrations must be sealed, walls, floors and ceilings must be water resistant and vacuum lines must be protected by traps or HEPA filters.

6. BSL-4: Standard Microbiological Practices

   All BSL-1, 2 and 3 Plus: A Class A positive pressure suit must be worn for entry into the laboratory.

   a. BSL-4: Special Practices

      1) BSL-3 Plus: All liquid effluent and solid waste must be decontaminated prior to disposal.

      2) Personnel must enter through a changing room and must change into laboratory clothes to wear underneath the positive pressure suit.

      3) Supplies must enter the laboratory through double door autoclaves or fumigation chambers.

   b. Secondary Barrier – Facility Requirements: All BSL1, 2 and 3 plus: a dedicated supply, exhaust, vacuum and decontamination system, double door autoclaves, the walls, ceilings and floors must be sealed, the doors must
be interlocked, a communication system between the laboratory and the outside is needed, and there must be emergency breathing air, an emergency generator and an emergency exit.

7. Animal Biosafety Levels:

The recommendations detailed below describe four combinations of practices, safety equipment, and facilities for experiments with animals infected with agents that cause, or may cause, human infection. These four combinations, designated Animal Biosafety Levels (ABSL) 1-4, provide increasing levels of protection to personnel and to the environment, and are recommended as minimal standards for activities involving infected laboratory animals. The four ABSLs describe animal facilities and practices applicable to work with animals infected with agents assigned to Biosafety Levels 1-4, respectively.

a. Animal Biosafety Level 1 (ABSL-1) is suitable for work involving well characterized agents that are not known to cause disease in healthy adult humans, and that are of minimal potential hazard to laboratory personnel and the environment.

b. Animal Biosafety Level 2 involves practices for work with those agents associated with human disease. It addresses hazards from ingestion as well as from percutaneous and mucous membrane exposure. ABSL-2 builds upon the practices, procedures, containment equipment, and facility requirements of ABSL-1.

c. Animal Biosafety Level 3 involves practices suitable for work with animals infected with indigenous or exotic agents that present the potential of aerosol transmission and of causing serious or potentially lethal disease. ABSL-3 builds upon the standard practices, procedures, containment equipment, and facility requirements of ABSL-2.

d. Animal Biosafety Level 4 involves practices suitable for addressing dangerous or exotic agents that pose high risk of life threatening disease, aerosol transmission, or related agents with unknown risk of transmission. ABSL-4 builds upon the standard practices, procedures, containment equipment, and facility requirements of ABSL-3. Procedures must be developed locally to address specific operations of the Class III cabinet line or the suit laboratory.

8. ABSL’s mimic BSL’s except for:

a. ABSL-1 must have self-closing and lockable doors, the interior surfaces must be water resistant, windows are not recommended, the floor drain traps must be filled with water and disinfectant, the air cannot be recirculated and the laboratory air pressure must be negative to the hallway.

b. ABSL-2 should have a mechanical cage washer which operates at 180°F, an autoclave must be available within the facility, and a hand washing sink must be in the room.

c. ABSL-3 must be physically separated from access corridors, with self-closing double-door access, preferably interlocked or alarmed, and windows and penetrations must be sealed.
d. ABSL-4 should have two workers in the laboratory when working with infected animals and the cages must be autoclaved or decontaminated before being cleaned.

9. There are also 4 biosafety levels for plants (BL1-4-P) listed in Appendix P of the NIH Guidelines. The requirements for practices and facilities are divided into Greenhouse Access levels (BL1-4-P) and standard laboratory plant biosafety levels (BL1-4-P).

   a. Greenhouse Access Level 1 (BL1-P) is a standard greenhouse with open windows and gravel walks permitted.

   b. Greenhouse Access Level 2 (BL2-P) is GAL-1 (BL1-P) plus screens over the openings and an autoclave available.

   c. Greenhouse Access Level 3 (BL3-P) is GAL-2 (BL2-P) plus an anteroom or head house, impervious bench tops and work surfaces, an autoclave inside the facility, an independent air supply with negative pressure, the exhaust is HEPA-filtered and a security fence or an equivalent form of security is present.

   d. Greenhouse Access Level 4 (BL4-P) is GAL-3 (BL3-P) plus the area is accessed through an airlock, there is a shower facility at all entrances and a dunk tank or fumigation chamber.

   e. Standard Laboratory Plant Biosafety Level 1 (BL1-P) has limited access, an entry log is maintained, a standard procedures manual must be used, experimental organisms must be inactivated, and a pest, rodent and weed control program must be in place.

   f. BL2-P is BL1-P plus biohazard signs in place where applicable, cages for small animals and procedures to minimize the escape of motile organisms.

   g. BL3-P is BL2-P plus access restricted to trained workers, equipment and supplies must be decontaminated, biohazards signs in place, efforts to minimize the formation of aerosols must be made, the surfaces of secondary containers used to take live organisms out of the laboratory must be decontaminated and a written record of accidents must maintained. Special clothing must be worn in the laboratory and the clothing must be decontaminated prior to laundering.

   h. BL4-P is BL3-P plus an entry/exit log must be strictly maintained, personnel must shower and change into special clothing upon entry and exit, all experimental materials and clothing must be decontaminated prior to removal and all accidents must be reported immediately.

10. There are BSL’s for large scale work. Large scale work is defined as any work involving ten or more liters. Prior to working at large scale at UTSA the Institutional Biosafety Officer must be consulted and the IBC must approve the protocols.

E. Hazards of tissue culture—human and primate tissue:

Work with human and non-human primate tissues and cell lines must be carried out at BSL-2. These tissues and cell lines can harbor human pathogens and required
precautions when working with them. They can also become contaminated with pathogens in the laboratory. Questions about individual cases should be addressed to the Biosafety Officer.

Tissue cultures from other animals can contain infectious agents, such as oncogenic viruses. Precautions should always be taken when working with tissue culture.

Fixed tissue is much less likely to contain infectious agents but as an example, prions can remain active after the fixation process.

F. IBC—Institutional Biosafety Committee

The UTSA Institutional Biosafety Committee is a registered committee with the National Institutes of Health. This Committee approves research protocols that involve infectious agents, recombinant DNA and the use of tissue isolated from vertebrates. The IBC is composed of UTSA research faculty, representatives from the UTSA Office of Environmental Health, Safety and Risk Management and community members outside the University. The committee meets the first Wednesday of each month, for applications to be considered they must be submitted by the 15th of the previous month.

1. Committee Charge

The charge of the Committee is to formulate and implement procedures to assure the University’s compliance with all federal regulations for the construction, handling and disposal of recombinant molecules, organisms and viruses containing recombinant DNA molecules and other biologically hazardous organisms and toxins at UTSA; to review and exercise approval authority of all proposals for grants and contracts that involve recombinant DNA molecules and other biologically hazardous organisms and toxins; to monitor all projects that involve recombinant DNA molecules and other biologically hazardous organisms and toxins; and to maintain the required records of the review, approval and monitoring of the use and disposal of recombinant DNA molecules and other biologically hazardous organisms and toxins.

2. Application to IBC for Approval

The application for submitting research to the IBC for approval can be found online at the IBC website http://research.utsa.edu/oric/. Only fill out the portion of the application pertaining to your research. The application should be submitted by the 15th of the month for review at the next month’s IBC meeting. For more information use the website to contact the IBC chair or the Institutional Biosafety Officer.

G. Bloodborne Pathogens Policies and Procedures

Laws and Regulations

Work with materials known or suspected to contain Bloodborne Pathogens (BBP’s) is regulated by both the federal and state governments. Federally OSHA regulates work with BBP’s through 29 CFR 1910.1030, the Bloodborne Pathogens Rule which became final in December 1991. In the state of Texas the Texas Department of State Health Services regulates work through 25 TAC Part 1
Chapter 96, Bloodborne Pathogen Control, which became final September 1, 2000. Disposal of waste which contains or could contain BBP’s is regulated by the Texas Commission on Environmental Quality through 30 TAC Chapter 330, 1201-1221, the Regulated Medical Waste Rule.

Both the federal and state regulations require annual training on BBP’s and a BBP Exposure Control Plan. UTSA’s Bloodborne Pathogens Exposure Control Plan can be found at the EHSRM website http://utsa.edu/safety/#/safetymanuals

H. Signage

Laboratories working with biohazardous materials must have door signage indicating these materials are used in the room. Laboratories working with BSL-2 and BSL-3 agents must have a sign indicating the specific agents used and any special precautions for entering as well as the biohazard symbol. Freezers, refrigerators and any equipment where agents are used or stored must be labeled with the biohazard symbol. Signs and labels are provided and/or posted by the Institutional Biosafety Officer or EHSRM Laboratory Safety Division personnel.

I. Select Agents

Select agents are biological agents or toxins that could pose a severe threat to public health and safety; to animal or plant health; or animal or plant products. A current listing of select agents and toxins can be found at the CDC website under the Select Agent Program. The most current list at the time of this plan’s update is available in Appendix III.

It is illegal to possess select agents without registration with the federal government. If you have any select agents which have not been registered, contact the Laboratory Safety Manager (LSM) immediately. If you plan to begin work with select agents, the application process should be started as early as possible. The process for approval for select agent work can take several months and requires additional security clearance procedures and approval of the Responsible Official for the UTSA Select Agent Program. Contact the LSM as soon as possible to begin the process.

J. Decontamination methods

There are different levels of “decontamination”.

1. Sterilization is a method which destroys all microbial life, including bacterial spores. Autoclaving (dry or steam) is one method of sterilization.

2. Disinfection is a method which reduces all forms of disease causing organisms on inanimate surfaces.

3. Decontamination is a method which reduces the numbers of organisms to acceptable levels.

4. Antisepsis is a method which reduces the number of organisms on living tissues.
5. The different decontamination methods which can be used for cleaning up biological spills, work area clean up and for equipment or glassware clean up are heat, radiation and chemicals. Heating involves dry or steam autoclaving which is sterilization method. Radiation in the form of ultraviolet light (UV) has limited effectiveness and should not be used as the only decontamination method. Chemicals have varying degrees of effectiveness according to the biological agent involved. Phenolics are tuberculocidal, but present a physical and health hazard. Aldehydes are sterilants, but present a health hazard and have limitations on surfaces. Halogens such as chlorine and iodine are tuberculocidal but, present health hazards and are unstable. For example bleach solution must be made fresh daily. Alcohols have a low level of effectiveness on surfaces but, are tuberculocidal as a soak. Quaternary Ammonium Compounds have a low level of effectiveness. Peroxygen Compounds are high to intermediate level in effectiveness (sporocidal), but can be costly to use.

K. Transportation and Shipping

Biological agents which are shipped fall into two categories: category A – Infectious Substances (UN 2814 or UN 2900) and category B - Biological Substances (UN 3373). There are various agencies and regulations to comply with when shipping. Entities involved in shipping include the USDA Animal and Plant Health and Inspection Service (APHIS), the Department of Transportation (DOT), the United States Postal Service (USPS), the Federal Aviation Administration (FAA), the International Air Transport Association (IATA) and the International Civil Aviation Organization (ICAO). Import/export permits are sometimes required, even within the US for specific agents. Specific shipping training with periodic re-training is required every 2 years for IATA member regulated air shipments and 3 years for DOT regulated ground shipments. Shipping without training can result in high fines and additional sanctions against the University.

1. Packaging for shipping biological agents requires a primary container with a positive seal surrounded by enough absorbent material to completely contain a spill. Secondary packaging which is watertight and leak proof holds the primary container. An outer container which must pass specific performance tests completes the containers. Packaging labels are necessary and between the secondary and outer packaging there must be a list of contents, the shipper’s label, including name, address and phone. The shipper’s label must also be on the outer container.

2. Example Packaging label for a category A – Infectious Substance
VI. Personal protective equipment (PPE)

Personal protective equipment (PPE) is a device or clothing worn to help protect you from direct exposure to hazardous materials. Examples include safety glasses or goggles, laboratory coats or aprons, gloves, face shields and respirators. Remember, PPE only protects you if you use it properly.

A. Eye and face protection

Eye protection must meet standards for impact resistance, provide splash protection and should provide UV protection. Safety glasses with side shields usually provide adequate impact resistance with limited splash protection. Chemical splash goggles (with no perforations around the goggles) provide adequate impact resistance and splash protection and limited vapor protection and therefore provide the best all-around eye protection. Vapor resistant goggles are available if needed. In addition to protective eyewear, face shields or freestanding shields should be used in situations where implosion or explosion may occur. Follow these guidelines for effective eye and face protection:

1. Wear protective eyewear at all times in the laboratory.

2. Wear chemical splash goggles for maximum protection, especially if you wear corrective lenses (glasses or contacts).

B. Hand protection

Gloves protect your skin from the biological agents you work with. Disposable latex gloves protect against water, dirt and microorganisms. Due to latex allergies or the possibility of developing such allergies, gloves made for other materials should be available in the laboratory. Information is available online, from the manufacturer or from EHSRM on the different types of gloves available and under
which circumstances they should be used. Follow these guidelines for effective hand protection.

1. Wear gloves that provide the greatest protection from the biological agents with which you are working.

2. Wash your hands promptly after removing protective gloves to avoid exposure due to microscopic holes, tears, or accidental contact with the outside of the gloves when removing them.

3. Remove gloves when handling common laboratory items (telephones, doorknobs, etc.) to prevent their contamination.

C. Body protection

The most common form of body protection in the laboratory is the laboratory coat. Laboratory coats protect your skin and clothes in the event of a spill or a splash. Follow these guidelines for effective body protection.

1. Protective clothing should be easily removable and free from rips or tears.

2. Wear your laboratory coat or apron only in the lab to prevent the potential spread of contamination.

3. Laboratory clothes should not be taken home to launder.

4. The following are not to be worn in laboratories: high-heeled or open-toed shoes, sandals or woven shoes, shorts or miniskirts, excessive jewelry.

D. Respiratory protection

Respiratory protection in the laboratory is normally provided by engineering controls such as the ventilation system and the biological safety cabinet(s). When a higher level of respiratory protection is required, an N-95 or PAPR (Powered Air Purifying Respirator) can be used. Contact EHSRM for assistance in selecting the correct respirator. Medical assessment, fit testing and training on proper use and storage are necessary prior to using a respirator. Follow these guidelines for effective respiratory protection:

1. Do not use a respirator unless you have been trained to do so and have undergone a medical evaluation.

2. If you are wearing a respirator, be sure appropriate fit-testing has been performed.

3. Properly store a respirator to prevent continued contamination.

VII. Laboratory equipment

A general understanding of laboratory equipment and how it works is essential to work safely in the laboratory.

A. Biological safety cabinets
Biological safety cabinets (BSCs) are among the most effective and most commonly used primary containment devices in laboratories working with infectious agents. The BSC is designed to capture and contain infectious particulates or aerosols generated within the BSC and exhaust them through a HEPA filter. Since HEPA filters are ineffective against volatile chemicals; therefore, volatile chemicals should not be used in BSCs.

Most BSCs recirculate 30 or 70% of their air within the cabinet. During this recirculation, the air passes over motors and wiring which are incompatible with a flammable atmosphere. For this reason flammables are not recommended for use in BSCs. Open flames can be problematic as well. The accidental release of flammable gas into the ventilation system of a BSC can result in a fire. Heat buildup from the use of a flame in a BSC can damage the HEPA filter, releasing infectious agents into the laboratory or the environment. Also important in some work is the fact that flames in BSCs will disrupt the laminar flow of the air and can cause contamination problems within the cabinet or in the laboratory.

For information on alternatives to flames in BSC’s contact the Institutional Biosafety Officer. NEVER have a BSC connected to natural gas lines without first gaining permission from the Institutional Biosafety Officer.

BSC’s must be installed and certified (annually) by a certified professional. EHSRM arranges the annual certification. BSC’s which have been moved must be recertified before use. Contact EHSRM to arrange recertification.

1. Classes of BSC’s

   a. Class I BSCs - These offer HEPA-filtered exhaust air; however, the supply air is not HEPA-filtered (“dirty room air” drawn inside), thus offering minimal protection to the user's hands and arms and vulnerable research materials inside the BSC. The Class I BSC is designed for general microbiological research with low- and moderate-risk agents (biosafety level 1 and 2 agents) and is useful for the containment of mixers, blenders and other equipment. Class I BSC’s are not routinely made.

2. Class II BSCs

   There are different types of Class II BSCs, but they all offer HEPA-filtered supply and exhaust air. This BSC protects the user, environment and research material and is suitable for work with moderate- to high-risk agents (biosafety level 2 and 3 agents). Class II BSCs are the most commonly used.

   a. Class II Type A1 cabinets were previously called Type A. This type of BSC recirculates 70% of the air and exhausts 30%. It has a positive pressure plenum. The Type A2 resembles the Type A1, but the positive pressure plenum is surrounded by negative pressure plenum. This cabinet was previously known as the Type B3. It can be connected to building exhaust with a thimble unit.

   b. The Class II Type B BSC is hard-ducted to an external exhaust system. The Type B1 recirculates 30% of the air and exhausts 70%. The cabinet has a positive pressure plenum with all contaminated air contained within a
negative pressure plenum. The supply blower is interlocked to prevent it from operating when the exhaust is insufficient. Emergency power can be connected to the exhaust blower to prevent the blower from shutting down.

c. The Class II Type B2 BSC is a total exhaust cabinet. Its blower is interlocked. Contaminated air is contained in positive pressure plenums enclosed in a negative pressure plenum. This is the only BSC in which small amounts of toxic chemicals, volatiles, and radionuclides (may need charcoal filters) may be used.

3. Class III BSCs

Often referred to as “glove boxes,” these gas-tight BSCs are under negative pressure. All work in the cabinet is done through rubber gloves attached to entry portals. The exhaust is double HEPA filtered and/or incinerated. Materials must enter through a sealed airlock and exit through an autoclave or dunk tank. Class III BSCs offer the highest level of protection and are suitable for work with extremely high risk agents (biosafety level 4).

4. Cabinets known as clean air centers, laminar flow clean air stations, laminar flow benches, aseptic work stations or horizontal/vertical flow benches function differently from BSC’s. They deliver HEPA filtered air across a bench top. This provides product protection and can be used with sterile equipment, for the assembly of equipment and preparation of sterile media. The air blows towards the worker. This can cause exposure to workers if improperly used with infectious materials. Reverse-flow cabinets of this type pull air from the front of cabinet through a pre-filter and HEPA filter at the rear. It can be used for cage changes, but PPE must be worn. These cabinets are not for work with biohazards since there is no containment.

5. Proper techniques for working in BSC’s:

   a. Always enter straight into cabinet with no sweeping motions.

   b. Place materials well within cabinet.

   c. Place the discard pan within cabinet.

   d. Watch for disruptions of the laminar flow.

   e. Decontaminate materials before removal from the cabinet.

   f. Protect vacuum lines with HEPA filters or traps.

B. Compressed Gas Cylinders

Compressed gas cylinders can present a dual hazard in the laboratory because the contents are under pressure and may contain hazardous materials, such as flammables, corrosives or toxics.

Follow these guidelines for proper use of compressed gas cylinders:

1. Compressed gas cylinders, empty or full, must be chained in place or otherwise secured at all times.
2. Cylinder caps must be in place except when the cylinder is in use.

3. Do not transport gas cylinders without the cylinder cap in place and an appropriate dolly with a securing strap.

4. Cylinder and delivery valves should be closed when not in use (especially true for toxic, flammable or corrosive gases).

5. Highly toxic, corrosive, and reactive gases present greater degrees of hazard. Work with these gases might require special containment, PPE, ventilation, piping systems, or alarm systems. Prior to ordering or working with these types of gases contact EHSRM for a risk assessment and determination of requirements.

6. Liquid nitrogen or any other liquefied gas can present additional hazards for handling and storage. Details on proper handling and storage can be found in the Chemical Safety Plan.

C. Centrifuges

Improper centrifuge use can result in the generation and release of hazardous aerosols. Centrifuges present a contamination problem when tubes break and the contents are released.

Follow these guidelines for proper centrifuge use:

1. Make sure the lid is on and secured before operating the centrifuge and remains secured until the centrifuge has come to a complete stop.

2. Always balance the load in the centrifuge. If you are not filling the entire centrifuge rack, position the tubes opposite one another. If you have an odd number of samples, use an empty tube with enough water to be of equivalent weight.

3. If vibration occurs, stop the centrifuge and check the load balances. Never operate an unbalanced centrifuge; this could result in breaking the centrifuge tube(s) and generating hazardous aerosols. Also, unbalanced rotors have the potential to become projectiles.

4. Keep the rotors and buckets clean, and promptly clean breakages or spills.

5. Ensure that the proper rotor is used for the centrifuge or the conditions of centrifugation.

D. Refrigerators

Follow these guidelines for proper laboratory refrigerator use:

1. Never place food or beverages in a refrigerator where biohazardous materials are stored.

2. Refrigerators containing biohazardous materials must be labeled “biohazard” with the appropriate biohazard symbol in black on an orange-red background.

E. Autoclaves
Autoclaves operate at high temperature and pressure and can present a physical hazard if not operated properly.

Follow these guidelines for proper autoclave use:

1. Users must be trained on operation prior to operating any autoclave.
2. Ensure cycles are completed and set correctly for the agent or sterilization will not be achieved.
3. Functionality tests must be performed periodically and recorded to confirm the autoclave is functioning properly.
4. Logs must be kept for each use of the autoclave.

VIII. Waste management
For information on biological waste disposal, refer to the UTSA Biological Waste Management Plan.

IX. Training
A. Researcher Biological Safety and Bloodborne Pathogens (SA 483)

This is a course on basic biological safety and bloodborne pathogens and is designed for persons working in biological research laboratories at UTSA. This course includes information about the regulations and what UTSA is doing to comply; principles/ concepts of biosafety, agent classes including bloodborne pathogens and safety levels, procedures and equipment that prevent exposure including engineering controls and personal protective equipment; sharps precautions, and clean-up procedures. New Employees (including transfers) are required to attend this training prior to initial assignment to duties that place them at risk of exposure to infectious agents. Current employees working with rDNA or biological agents shall attend.

B. Annual Refresher Researcher Biological Safety and Bloodborne Pathogens (SA 483r)

This course is designed to meet mandated requirements for an annual training refresher.

C. Hazard Communication and Laboratory Safety (SA 443)

Hazard Communication training is mandated by both the federal and state governments. If you will be exposed to hazardous chemicals within your work area, you must attend Hazard Communication and Laboratory Safety training. Hazardous chemicals are defined as chemicals which have a physical or health effect.

D. Hazardous Waste Generator (SA 443)

Hazardous Waste Generator training covers chemical and biological waste disposal procedures in accordance with federal, state and local regulations.
Generators must understand the requirements for proper bulking, packaging, labeling and disposal of hazardous waste.

X. Emergency procedures and equipment

Due to the multiple hazards associated with laboratories, incidents are inevitable. Preparedness for emergencies is essential. A timely and efficient response can help minimize or avoid injury and damage to property. For a comprehensive discussion of UTSA emergency procedures, including internal (fire, bomb threat) and external (tornado, flooding) emergencies, see UTSA’s Emergency Response Plan.

A. Biological spills

Response to biological spills must be thorough and prompt to prevent further injury or contamination.

Each laboratory should design its own response plan based on its unique hazards and the location of the laboratory, in conjunction with the following general guidelines:

1. Notify the people in the immediate area and, if necessary, evacuate the laboratory. The decision to evacuate is a judgment call based on the properties and hazards of the spilled biological agent. If biological aerosols result from the spill, evacuation should follow. Contact Facilities Operations immediately; tell them to shut off air handlers to prevent the spread of hazardous aerosols if they have escaped from the laboratory’s containment.

2. Always attend to injured people before attending to the spill. Skin areas splashed by biologicals should be rinsed with water for at least 15 minutes in a sink, emergency shower or eyewash as appropriate. After thorough rinsing, seek medical help. Be sure to have the identity of the biological agent and other information, if possible, available for the medical help.

3. Try to contain the spill to keep it from spreading. Contact EHSRM to advise or assist in the containment, disinfection and cleanup of the spilled biological agent. Do not attempt to clean the spill without proper spill-control supplies or equipment.

4. If the spill or release is likely to affect other facilities within the building or campus, contact the UTSA Police Department. UTSA PD can be reached in an emergency at X911 on a campus phone and 458-4911 on an outside phone, such as a cell phone.

B. Emergency equipment

Laboratory emergency equipment includes emergency showers, eyewashes and fire extinguishers. Staff in laboratories that do not have their own emergency shower and eyewash station should know where the closest one is located.

1. Showers

An emergency shower is used to decontaminate someone who has been exposed to biological agents or chemicals.
a. Remove clothing, jewelry and shoes while standing under the shower. These items trap agents against the skin and will prevent proper cleaning if not removed.

b. Remain under the shower for at least 15 minutes to ensure adequate flushing of exposed areas.

c. Seek medical attention.

d. If the shower does not have a drain, promptly clean up the water to prevent slip hazards adding the appropriate decontamination agent.

e. Always keep the area under an emergency shower unobstructed.

2. Eyewashes

a. If biological agents are splashed into your eyes, locate the nearest eyewash station and hold your top and bottom eyelids open and flush with water continuously for at least 15 minutes. Move the eye up and down and sideways to wash thoroughly behind the eyeball where agents could be trapped.

b. Seek medical attention.

c. Always flush your eyes immediately if biological agents are splashed into them. Immediate action may prevent an infection.

d. Continuous-flow eyewashes are preferred over the portable, squeeze-bottle type, whose disadvantages include an insufficient supply of water (not 15 minutes’ worth) and easy contamination with microorganisms. Squeeze-bottle and non-plumbed eyewashes are not allowed at UTSA.

e. To ensure a clean supply of water in the eyewash, operate it weekly to flush out any impurities.

XI. Laboratory Deactivation and Equipment Disposal

A. Equipment Disposal Procedure: See Appendix III for details.

Equipment to be disposed of should be wiped down with an appropriate disinfectant solution such as a bleach solution or an ethanol solution.

Once the equipment has been cleaned, EHSRM should be contacted to check the equipment and place proper signage on it stating that it has been reviewed and is ready to be removed.

Laboratory personnel should then contact the Inventory and Surplus Department to have the equipment removed from the laboratory.

B. Laboratory Deactivation Procedure: See Appendix III for details.

EHSRM should be contacted before a laboratory deactivation begins. Pertinent personnel from EHSRM will come by the laboratory to review what items need to be dealt with. For instance, laboratory safety personnel will review what areas
and equipment need to cleaned due to possible biological or chemical contamination.

The Radiation Safety Officer (RSO), Radiation & Laser Safety Coordinator or other EHSRM Laboratory Safety Division personnel will review for possible radiological contamination and determine what measures must be taken to deal with it. Chemical and biological wastes will also be reviewed by the appropriate personnel. Once areas and equipment have been properly cleaned using a disinfectant or appropriate solvent, EHSRM personnel will need to review to determine if all cleaning has been done properly. Any equipment will be labeled as ready to move, repair, or for disposal. The area or laboratory itself will be labeled as appropriately decontaminated and ready for Housekeeping staff to do routine cleaning to prepare for its future occupants.

XII. References


1. Appendix A - Primary Containment for Biohazards: Selection, Installation and Use of Biological Safety Cabinets.

2. Biological Safety in the Laboratory: A Guide for Biological Safety and Handling Biological Agents. The University of Texas Health Science Center at San Antonio, Department of Institutional Safety, 1995.


XIII. Appendices
Appendix I Classification of Infectious Agents on the Basis of Hazard
Appendix II List of Select Agents and Toxins
Appendix III Equipment and Laboratory Clean out/Clearance Procedures
Appendix I Classification of infectious agents on the basis of hazard

1. **Risk Group 1 agents**

   All bacterial, parasitic, fungal, viral, rickettsial and chlamydial agents not associated with disease in adults. Agents not included in higher classes are not by default in Risk Group 1.

2. **Risk Group 2 agents**

   **Bacterial agents**
   
<table>
<thead>
<tr>
<th>Agent</th>
<th>K1 antigen</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Acinetobacter calcoaceticus</em></td>
<td><em>Klebsiella</em>-spp. and serotypes</td>
</tr>
<tr>
<td><em>Actinobacillus</em>-spp.</td>
<td><em>Legionella pneumophila</em></td>
</tr>
<tr>
<td><em>aeromonas hydrofoil</em></td>
<td><em>Leptospira interrogans</em>-spp</td>
</tr>
<tr>
<td><em>Arizona hinshawii</em>-all serotypes</td>
<td><em>Listeria</em>-spp.</td>
</tr>
<tr>
<td><em>Bacillus anthracis</em></td>
<td><em>Moraxella</em>-spp.</td>
</tr>
<tr>
<td><em>Bordetella</em>-spp.</td>
<td><em>Mycobacterium</em>-spp. (except those listed in Risk Group 3)</td>
</tr>
<tr>
<td><em>Borrelia recurrentis, B. vincentii</em></td>
<td><em>Neissaria gonorrhoea, N. meningitides</em></td>
</tr>
<tr>
<td><em>Campylobacter fetus</em> in Risk Group 3</td>
<td><em>Pasteurella</em>-spp. (except those listed)</td>
</tr>
<tr>
<td><em>Campylobacter jejuni</em></td>
<td><em>Salmonella</em>-spp. and all serotypes</td>
</tr>
<tr>
<td><em>Chlamydia psittaci</em></td>
<td><em>Shigella</em>-spp. and all serotypes</td>
</tr>
<tr>
<td><em>Chlamydia trachomatis</em></td>
<td><em>Sphaerophorus necrophorus</em></td>
</tr>
<tr>
<td><em>Clostridium botulinum</em></td>
<td><em>Staphylococcus aureus</em></td>
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<tr>
<td><em>Cl. chuvoei</em></td>
<td><em>Streptobacillus moniliformis</em></td>
</tr>
<tr>
<td><em>Cl. haemolyticum</em></td>
<td><em>Streptococcus pneumoniae</em></td>
</tr>
<tr>
<td><em>Cl. histolyticum</em></td>
<td><em>S. pyogenes</em></td>
</tr>
<tr>
<td><em>Cl. novyi, Cl. septicum, Cl. tetani</em></td>
<td><em>Treponema cratum</em></td>
</tr>
<tr>
<td><em>Corynebacterium diptheriae, C. equi</em></td>
<td><em>T. pallidum</em></td>
</tr>
<tr>
<td><em>C. haemolyticum, C. pseudotuberculosis</em></td>
<td><em>T. pertenue</em></td>
</tr>
<tr>
<td><em>C. pyogenes, C. renale</em></td>
<td><em>V. parahaemolyticus</em></td>
</tr>
<tr>
<td><em>Edwardsiella tarda</em></td>
<td><em>Vibrio cholerae</em></td>
</tr>
<tr>
<td><em>Erysipelothrix insidiosa</em></td>
<td><em>Yersinia enterocolitica</em></td>
</tr>
<tr>
<td><em>Erysipelothrix rhusiopathiae</em></td>
<td></td>
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<tr>
<td><em>Escherichia coli</em></td>
<td></td>
</tr>
<tr>
<td><em>Helicobacter pylori</em></td>
<td></td>
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<tr>
<td><em>Listeria monocytogenes</em></td>
<td></td>
</tr>
<tr>
<td><em>Mycobacterium</em></td>
<td></td>
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<tr>
<td><em>Neisseria</em></td>
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<tr>
<td><em>Pseudomonas</em></td>
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<tr>
<td><em>Salmonella</em></td>
<td></td>
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<tr>
<td><em>Staphylococcus</em></td>
<td></td>
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<tr>
<td><em>Streptococcus</em></td>
<td></td>
</tr>
<tr>
<td><em>Treponema pallidum</em></td>
<td></td>
</tr>
</tbody>
</table>
*Escherichia coli*—all enteropathogenic, entero-toxigenic, enteroinvasive and stains bearing

*Haemophilus ducreyi*,  
*H. influenzae*

Mycoplasma-spp. (except Mycoplasma mycoides and M. agalactiae, which are forbidden)

**Fungal agents**

Actinomycetales (including Actinomyces spp. and Arachnia propionica)

*Blastomyces dermatitidis*

*Cryptococcus neoformans*

*Paracoccidioides brasiliensis*

**Parasitic agents**

*Entamoeba histolytica*

*Leishmania*-spp.

*Naegleria gruberi, N. fowleri*

*Schistosoma mansoni*

*Toxoplasma gondii*

*Toxocara canis*

*Trichinella spiralis*

*Trypanosoma cruzi*

**Viral, rickettsial and chlamydial agents**

*Adenoviruses*, human—all types

*Cache Valley virus*

*Corona viruses*

*Coxsackie A and B viruses*

*Cytomegaloviruses*

*Echoviruses*—all types

*Encephalomyocarditis virus (EMC)*

*Flanders virus*

*Hart Park virus*
Hepatitis-associated antigen material

Herpesvirus-associated antigen material

Herpesviruses (except Herpesvirus simiae—Monkey B virus—which is Risk Group 4)

hTLV I/II

Human Immunodeficiency virus (except large volumes or high concentrations that require BL3)

Influenza viruses—all types except A/PR8/34, which is Risk Group 1

Langat virus

Measles virus

Mumps virus

Parainfluenza viruses—all types except

Parainfluenza virus 4, SF 4 strain, which is Risk Group 1

Polio viruses—all types, wild and attenuated

Pox viruses—all types, except Alastrim, Smallpox and Whitepox, which are forbidden; and Monkey pox, which, depending on the experiment, is Risk Group 3 or 4

Rabies virus—all strains except Rabies street virus, which is Risk Group 3 or 4

Reoviruses—all types

Respiratory syncytial virus

Rhinoviruses—all types

Rochalimaea vinsonii

Rubella virus

Simian viruses—all types except Herpesvirus simiae (Monkey B virus) and Marburg virus, which are Risk Group 4

Sindbis virus

Tensaw virus

Turlock virus

Vaccinia virus

Varicella virus
Vesicular stomatitis virus

Yellow fever virus, 17d vaccine strain

3. Risk Group 3 agents

Bacterial agents

Bartonella -spp.
Brucella -spp.

Francisella tularensis

Mycobacterium avium complex, M. bovis, M. tuberculosis

Pasteurella multocida type B (“buffalo” and other foreign virulent strains)

Yersinia pestis

Fungal agents

Coccidioides immitis

Histoplasma capsulatum

Histoplasma capsulatum var duboisii

Parasitic agents

None

Viral, rickettsial, and chlamydial agents

Arboviruses—all strains except those in Risk Group 2 and 4

Coxiella burnetii

Ehrlichia-spp.

Lymphocytic choriomeningitis virus (LMC)

Monkey pox virus, when used in vitro

Rabies street virus

Rickettsia-spp. (except R. ruminantium)

West Nile and Semliki Forrest viruses, depending on conditions of use and geographical location of the laboratory

Yellow fever virus, wild, when used in vitro

4. Risk Group 4 agents

Bacterial, fungal and parasitic agents
None

**Viral, rickettsial and chlamydial agents**

*Ebola fever virus*

*Hemorrhagic fever agents*, including Crimean hemorrhagic fever, Congo, Junin and Machupo viruses

*Herpesvirus simiae (Monkey B virus)*

*Lassa fever virus (Mastomys natalensis)*

*Marburg virus (Cercopithecus spp.)*

*Monkey pox*, when used for transmission or animal inoculation experiments

*Tick-borne encephalitis virus complex*, including Russian spring-summer encephalitis, Kyasanur forest disease, *Omsk hemorrhagic fever* and *Central European encephalitis viruses*

*Venezuelan equine encephalitis virus*—epidemic strains, when used for transmission or animal inoculation experiments

*Yellow fever virus*—wild, when used for transmission or animal inoculation experiments

5. **Low-risk oncogenic viruses**

*AD7-SV40*

*Adenovirus*

*Avian leukosis*

*Bovine leukemia*

*Bovine papilloma*

*CELO*

*Dog sarcoma*

*Guinea pig herpes*

*Hamster leukemia*

*HTLV I/II*

*Lucke (frog)*

*Marek’s*
Mason-Pfizer monkey virus
Mouse mammary tumor
Murine leukemia
Murine sarcoma
Polyoma
Rat leukemia
Rat mammary tumor
Rous sarcoma
Shope fibroma
Shope papilloma
SV-40
Moderate-risk oncogenic viruses
Ad2-SV40
EBV
FeLV
FeSV
GaLV
HV Ateles
HV Saimiri
SSV-1
Yaba
Appendix II. List of Select Agents and Toxins

HHS AND USDA SELECT AGENTS AND TOXINS

HHS SELECT AGENTS AND TOXINS

Abrin
Botulinum neurotoxins
Botulinum neurotoxin producing species of *Clostridium*
Cercopithecine herpesvirus 1 (Herpes B virus)
*Clostridium perfringens* epsilon toxin
*Coccidioides posadasii/Coccidioides immitis*
Conotoxins
*Coxiella burnetii*
Crimean-Congo haemorrhagic fever virus
Diacetoxyscirpenol
Eastern Equine Encephalitis virus
Ebola virus
*Francisella tularensis*
Lassa fever virus
Marburg virus
Monkeypox virus
Reconstructed replication competent forms of the 1918 pandemic influenza virus containing any portion of the coding regions of all eight gene segments
(Reconstructed1918 Influenza virus)
Ricin
*Rickettsia prowazekii*
*Rickettsia rickettsii*
Saxitoxin
Shiga-like ribosome inactivating proteins
Shigatoxin
South American Haemorrhagic Fever viruses
  *Flexal*
  *Guanarito*
  *Junin*
  *Machupo*
  *Sabia*
Staphylococcal enterotoxins
T-2 toxin
Tetrodoxin
Tick-borne encephalitis complex (flavi) viruses
  *Central European Tick-borne encephalitis*
  *Far Eastern Tick-borne encephalitis*
  *Kysanur Forest disease*
  *Omsk Hemorrhagic Fever*
  *Russian Spring and Summer encephalitis*
Variola major virus (Smallpox virus)
Variola minor virus (Alastrim)
*Yersinia pestis*

OVERLAP SELECT AGENTS AND TOXINS

Bacillus anthracis
*Brucella abortus*
*Brucella melitensis*
*Brucella suis*
*Burkholderia mallei* (formerly *Pseudomonas mallei*)
*Burkholderia pseudomallei* (formerly *Pseudomonas pseudomallei*)
Hendra virus
Nipah virus
Rift Valley fever virus
Venezuelan Equine Encephalitis virus

USDA SELECT AGENTS AND TOXINS

African horse sickness virus
African swine fever virus
Akabane virus
Avian influenza virus (highly pathogenic)
Bluetongue virus (exotic)
Bovine spongiform encephalopathy agent
Camel pox virus
Classical swine fever virus
*Ehrlichia ruminantium* (Heartwater)
Foot-and-mouth disease virus
Goat pox virus
Japanese encephalitis virus
Lumpy skin disease virus
Malignant catarrhal fever virus
  *(Alcelaphine herpesvirus type 1)*
Menangle virus
*Mycoplasma capricolum* subspecies *capripneumoniae*
  *contagious caprine pleuropneumonia)*
*Mycoplasma mycoides* subspecies *mycoides* small colony *(MmmSC)* (contagious bovine pleuropneumonia)
Peste des petits ruminants virus
Rinderpest virus
Sheep pox virus
Swine vesicular disease virus
Vesicular stomatitis virus (exotic): *Indiana subtypes VSV-IN2, VSV-IN3*
Virulent Newcastle disease virus

USDA PLANT PROTECTION AND QUARANTINE (PPQ)

SELECT AGENTS AND TOXINS

*Peronosclerospora philippinensis* *(Peronosclerospora sacchari)*
*Phoma glyciniola* (formerly *Pyrenochaeta glycines*)
*Ralstonia solanacearum* race 3, biovar 2
*Rathayibacter toxicus*
*Sclerophthora rayssiae var zeae*
*Synchytrium endobioticum*
*Xanthomonas oryzae*
*Xylella fastidiosa* (citrus variegated chlorosis strain)

1 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.
Appendix III. EQUIPMENT AND LABORATORY CLEAN OUT / CLEARANCE PROCEDURES

EQUIPMENT CLEAN OUT / CLEARANCE PROCEDURE

When equipment that is used with hazardous materials, requires repair or disposal laboratory personnel need to ensure the equipment is cleaned of all hazardous biological, chemical and radioactive materials. Environmental Health Safety and Risk Management (EHSRM) must certify equipment to be cleared of hazards prior to repair, shipping for repair, sending to surplus or disposing of equipment.

- All equipment, including chemical fume hoods, must be cleaned / decontaminated to remove any hazardous materials or residue including chemicals, potentially infectious biological agents, and radioactive materials.

  - For biological agents: An appropriate tuberculocidal grade disinfectant such as 1-10% dilution of household bleach (5.25-6% sodium hypochlorite solution) applied for a contact time of 10-20 minutes should be used. Alternately, a commercially available detergent-disinfectant solution such as Dispatch®, Clorox Clean-up®, Lysol IC®, etc. may be used following manufacturer’s instructions. Caution – most disinfectants are also corrosive – proper PPE should be worn (gloves & faceshield or goggles) and metal surfaces especially should be rinsed with water and wiped down after application.
  
  - For chemicals: An appropriate solvent for the chemical residues which may be present should be used, followed by a detergent cleaning.
  
  - For radioactive material areas: Wipe tests shall be completed prior to an appropriate detergent solution wipe down. If wipe tests confirm areas of contamination, then all decontamination materials must be kept for radioactive waste disposal. If applicable, final wipe tests shall be conducted to verify proper decontamination. All wipe tests and survey locations must be documented.

- Some equipment will need specialized cleaning/decontamination. For example, biological safety cabinets will need to be decontaminated with formaldehyde, vaporized hydrogen peroxide or other materials. Currently this type of decontamination is not done in-house. Contact the Laboratory Safety Division for more information.

- EHSRM must be contacted to remove any remaining waste biological agents or hazardous chemicals for disposal. Environmental Safety Division is the contact for this service.

- If radioactive materials were used in the equipment then Radiation Safety Personnel (RSP) must be contacted to clear the area. Once complete, RSP will complete and sign the radiation portion of the equipment clearance tag.

- For biological or chemical clearance of equipment, either the Environmental or Laboratory Safety Division can be contacted to do the review and fill out the equipment clearance tag.

- Once the equipment clearance tag has been signed and posted by pertinent EHSRM personnel, the equipment can be repaired or removed from the lab for service, surplus or disposal.

CONTACTS:

Environmental Protection & Construction Safety Division: X6698, X5808
Laboratory Safety Division: X6697, X5807, X6101
Radiation Safety Personnel: X6697, X6101

See [http://utsa.edu/safety/](http://utsa.edu/safety/) for more contact information.