Laboratory Safety Manual

Touro University California

In Conjunction with TUCA's Institutional Biosafety Committee

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Appendices

TUCA Institute Biosafety Committee
Contact Information

Athena W. Lin, Ph.D
Biosafety Officer (707) 638-5239
Provides information about TUCA policy on biosafety, clinical trials, Institutional Biosafety Committee, new guidelines from NIH Office of Biotechnology Activities and the Recombinant DNA Advisory Committee.
Provides information regarding biosafety considerations of using infectious agents in research, Institute Biosafety Application and Memorandum of Understanding and agreement review process, facilitates contact between scientists and CDC/NIH representatives, helps in implementing Federal and State guidelines.

Daniel Keppler, Ph.D
Chair of IBC
(707) 638-5956

Tamira Elul, Ph.D
Chair of Safety Committee
Member of IBC
(707)638-5453

Nathalie Garcia-Russell, Ph.D
Member of IBC
(707) 638-5917

Miriam Gochin, Ph.D
Member of IBC
(707) 638-5463

Alison McCormick, Ph.D
Member of IBC
(707) 638-5987

David Madden, Ph.D
Member of IBC
(707) 638-45248

Evan Hermel, Ph.D
Member of IBC

Vanishree Rajagopalan, Ph.D
Member of IBC
(707) 638-5928

EMERGENCIES (from a campus phone): ***9-911(police/fire)***
8-5804(Security)

Section I – BIOLOGICAL SAFETY

A. Introduction

The purpose of this Laboratory Safety Manual is to provide information concerning TUCA's Safety Policy and Procedures thereby promoting a safe working environment. Although the manual is primarily designed for compliance by the Institute's scientific, technical, and supportive staffs, all other employees must be knowledgeable about these safety policies and the procedures for implementation. In 1983, the Federal Occupational Safety and Health Administration (OSHA) set forth the Occupational Safety and Health Standard entitled, "Hazard Communication Standard" (29 CFR 1910.1200) and Laboratory Standards (29 CFR 1910.1450). These standards and similar existing state and local governmental ordinances have been commonly called the "Worker's Right to Know" laws which provide minimum standards that employers must adhere to for informing employees about occupational-related hazards in the work place.

On August 28, 1987, OSHA published the Final Rule (Standard) (See Section VI. 1.) which supersedes all State and Local Regulations regarding the use of toxic substances in laboratories.

The policies, regulations and procedures defined in this manual are one method of compliance with the Worker's Right to Know Laws. However, this manual has a much broader scope than occupational-related hazards. It is not just a means for the Institute to meet its obligation to inform its employees, but it is also a guide to follow in making the Institute a safer workplace. Accordingly, this manual covers a wide spectrum of safety precautions, ranging from daily housekeeping chores to procedures to follow in emergencies. It addresses the following five specific issues:

1. General Laboratory Safety
2. Biological Safety
3. Chemical Safety
4. Radiation Safety
5. Role and Responsibility of the Institute's Biosafety Committee and the Institute's Biosafety Officer.

It is the responsibility of each employee, student, volunteer and visiting faculty and staff to follow the rules of laboratory safety. It is the responsibility of each laboratory employee, particularly the Principal Investigator to read and understand the information contained in the Manual and to keep the Manual readily accessible for review and emergency usage. The Manual will be updated as new safety information or governmental regulations are obtained. TUCA reserves the right to delete, add or amend the contents of this Manual. Occupational hazard regulatory rules will undoubtedly continue to be changed. Accordingly, no representation can be made or responsibility undertaken by TUCA regarding the completeness, accuracy, or continuing validity of the contents of this Manual.
In the final analysis, each employee must assume his or her responsibility to work in a safe manner, thereby avoiding personal harm or endangering others.

Section II – RIGHT TO KNOW GUIDELINES

A. Declaration

> ***Note:*** *This section is required to be reviewed and signed upon acceptance of a position or before the employee begins working at TUCA.*

Biomedical research often requires the use of hazardous materials including radioisotopes, infectious agents, and hazardous chemicals. While working at TUCA, it is likely that you will be required to handle such materials. It will be your specific right and obligation to know, before using a hazardous material in an experiment, what is the nature of the material, its specific hazard and the proper procedures for its use.

Radioactive material use will be monitored by and under the control of TUCA's Radiation Safety Office.

Prior to utilizing any substance, each employee of TUCA has the right and obligation to be educated in the proper use and risks associated with the substance. If, as an employee of TUCA, you have any questions about any substance you work with, you should contact your Principal Investigator or the Safety Officer (chemical hazards), Biosafety Officer (biohazards). (*See TUCA Biosafety Committee Contact Information on Page 5)* for telephone numbers.

With your Right to Know come specific responsibilities for your protection and the protection of others. It is mandatory that all employees adhere to government and TUCA's guidelines and regulations in the use and disposal of any hazardous materials. In addition, all reasonable precautions to assure the safety of yourself and others must be taken.

If you are ever in doubt, have a problem with the use of any materials, or have a complaint about experiments done by others, the following procedures are to be followed:

1. Discuss the problem with your immediate supervisor.
2. If unsatisfied, discuss the problem with the Institute's Biosafety Officer.
3. If still unsatisfied, contact the Institute's Biosafety Committee Chair.

It is the policy of TUCA to provide a safe working environment for personnel and to provide documentation of policies and procedures which have been implemented to eliminate or reduce employee exposure to bloodborne pathogens (See Appendix A). Procedures have been developed to identify those individuals with occupational exposure to blood and other potentially infectious materials and provide them with training, protective equipment, hepatitis B vaccine, and post exposure follow-up in accordance with the OSHA Standard on Occupational Exposure to Bloodborne Pathogens (See 29 CFR Part 1910.1030) http://www.osha.gov/SLTC/bloodbornepathogens/standards.html and current recommendations from the Centers for Disease Control and Prevention (CDC). All research laboratory personnel who have any exposure to bloodborne pathogens should contact the Employee Health Clinic immediately. If it is after business hours, the employee should call the BSO or 911 if urgent.

B. Introduction to Universal and Standard Precautions:

Universal Precautions were developed specifically to prevent infections from bloodborne pathogens. Standard Precautions basically expands upon Universal Precautions by covering more body fluids and tissues. All human blood and certain body fluids are treated as if they are known to be infectious for HBV, HIV and other bloodborne pathogens. Universal Precautions are intended to prevent parenteral, mucous membrane, and non-intact skin exposures of workers to bloodborne pathogens. Universal Precautions apply to blood and body fluids containing visible blood, tissues, semen, vaginal secretions, cerebrospinal, synovial, pleural, peritoneal, pericardial, and amniotic fluids. Universal Precautions do not apply to feces, nasal secretions, sputum, sweat, tears, urine, vomit, or saliva unless they contain visible blood.

Section III – LABORATORY SAFETY TRAINING

A. Policy

As an integral part of TUCA, all laboratory personnel must attend the mandatory Chemical Hygiene Course concerning the Safety Policies and Procedures of TUCA.

Laboratory safety training will be obtained in accordance with TUCA's Safety Policies and Procedures.

The Principal Investigator in each research laboratory is responsible for ensuring that all laboratory activities under his or her control are conducted in a manner that presents the least possible hazard to employees and the surrounding community. The Principal Investigator must ensure that all safety policies and regulations are enforced and that necessary safety equipment is available in the laboratory. The Principal Investigator has the primary responsibility for the health and safety of all personnel under his/her jurisdiction including employees, students, guest scientists and visitors.

The Principal Investigator's responsibilities include:

1. Identification of hazards and assessment of the risks associated with operations.
2. Ensuring that laboratory personnel are aware of hazards and of the precautions they should take in carrying out their assigned tasks.
3. Selection of proper laboratory safety practices and engineering controls necessary to minimize personal injury or property damage.
4. Informing laboratory personnel of the rationale for selection of appropriate preventive medical practices, serologic monitoring, and immunization protocols in conjunction with Human Resources, Biosafety Committee, Biosafety Officer.
5. Providing instruction and training programs for personnel in the practices and techniques required for their assigned tasks and laboratory operations.

6. Maintaining the Laboratory Safety Manual.

7. Ensuring that necessary safety equipment is available in the laboratory, used when required, and adequately maintained.

8. Establishing and periodically reviewing emergency procedures for accidental spills and any overt exposure to hazardous substances in conjunction with the Institute Biosafety Committee and Safety Office.

9. Complying with all policies and procedures as outlined in this Manual.

Each employee and student working in a research laboratory has the following responsibilities:

1. Complying with the Institute's Safety Policies and Procedures.

2. Maintaining awareness of the risks associated with assigned duties.

3. Taking all necessary and appropriate safety precautions relevant to the performance of their duties.

4. Becoming familiar with emergency procedures prior to accidental spills, overt personal exposures, fire, etc.

5. Reporting unsafe conditions or practices to the Principal Investigator, Safety Officer, Research Administrator, or Chair of the Biosafety Committee.

6. Reporting all incidents resulting in injury or exposure to hazardous agents to employee's supervisor and Employee Health Services.

Health and safety awareness will be promoted among principal investigators, managers, supervisors, employees, students and others (visitors, contractors, community members) through orientation programs and regularly scheduled education and training sessions, as appropriate.

B. Laboratory Safety Training Curriculum

The Laboratory Safety Training curriculum shall include, but not be limited to, the following list of subject matter:

1. Introduction
    a. Personal Safety
    b. Safety of Non-Laboratory Personnel
    c. Safety Manuals
    d. Safety Committees
    e. Fire Safety/Emergency Preparedness

2. Chemical Safety
    a. Fire Safety
    b. The Right-to-Know Laws
    c. Chemical Handling, Transfer, and Storage

3. Biological Safe
    a. Hazards
    b. Procedures
    c. Restricted Access Areas
    d. Levels of Containment
    e. Infectious Agents (See Appendix B)
    f. Vaccination and Surveillance

4. Radiation Safety
    a. Radioactive Compounds
    b. Cesium Sources
    c. Radiation Producing Machines

5. Common Use Equipment
    a. Gamma and Scintillation Counters
    b. Lasers
    c. Liquid Nitrogen Freezers
    d. Autoclaves
    e. Central Deionized Water
    f. Equipment Repairs through Physical Plant
    g. Biological Safety Cabinets and Fume Hoods

6. Waste Management
    a. Infectious Waste
    b. Chemical Waste
    c. Radioactive Waste

7. Laser Safety
    a. Operation
    b. Protective Eyewear
    c. Posting
    d. Control Measures

All new personnel are mandated to attend a Chemical Hygiene Course. Dates and locations can be found on TUCA's webpage. Employees who have taken the Chemical Hygiene Course must renew their certification every two years.

C. Special Orientation for Biosafety Level 3

Specific orientation sessions will be held for all laboratory personnel whose work assignments require the use of the Institute's BSL 3. The orientation session must be held and

approval obtained prior to allowing the applicant admission to these facilities. Whenever possible, the orientation will be held on an individual basis and shall consist of the following steps:

1. Personal interview with the applicant by the Biosafety Officer for determining the applicant's knowledge and experience with indigenous or exotic agents.
2. Applicant will be given a copy of the Standard Operating Procedures.
3. Applicant will be required to sign an acknowledgment form.
4. Applicant will have their ID cards activated for entrance and exit to and from the laboratory.
5. Facility users are encouraged to participate in a voluntary surveillance program to monitor occupational exposure to infectious agents.
6. Applicants will work on projects approved by the Institute Biosafety Committee.

SECTION IV – GENERAL LABORATORY SAFETY

A. Housekeeping

Many safety and health problems can be avoided through observance of good housekeeping procedures including cleaning the work area and general maintenance of the laboratory. All unnecessary glassware and materials must be removed from bench tops after use to avoid clutter that may cause accidents. Floors should be kept free of boxes, instruments, and supplies by storing them properly.

B. Protective Clothing

A laboratory coat must be worn when in a laboratory while conducting experiments. Laboratory coats are worn to protect regular clothes from hazardous materials and should be removed whenever leaving the laboratory environment. Open-toed footwear should not be worn in laboratories at TUCA. As a further precautionary measure, disposable gloves, chemical aprons, respirators, goggles or eye shields must be used when appropriate. Furthermore, disposable gloves are not to be worn outside of the laboratories, i.e., gloves must never be worn in hallways, elevators, or public areas of the Institute. With exception when transporting hazardous materials, one hand must be gloved for protection, leaving the other hand ungloved to facilitate opening doors, pressing elevator buttons, etc.

C. Eating, Drinking, and Applying Cosmetics

Eating, drinking, food preparation and application of cosmetics in laboratories are forbidden. Lunches MUST NOT be stored in laboratory refrigerators.

D. Pipetting

Pipetting by mouth is forbidden. This regulation is very important. There are many alternatives to mouth pipetting. With a little practice, it is possible to work both quickly and accurately with mechanical devices. The only exception is mouse embryo transfer in the transgenic facility.

E. Needles and Pasteur Pipettes

Attempts to re-sheath needles or remove them from the syringe should be avoided as they result in accidents. Unsheathed needles must be carefully placed in the special sharps container (available from Central Supply) for disposal. The container must be sealed with tape and marked: “Autoclave and Discard” as specified in the Institute’s Policy and Procedure.

F. Broken Glassware

Recommended procedures:

1. Dispose of any damaged glassware.
2. Do not use cracked or chipped glassware.
3. When disposing of cracked, chipped, or broken glassware, use forceps and heavy gloves. Place disposable glassware in the appropriate containers located in the laboratory.

G. Gas Cylinders

All cylinders including empty cylinders must be firmly secured to the wall, bench or cart. Proper regulators must be used. Do not lubricate, modify or tamper with a cylinder or regulator valve.

H. Hazard Warnings

As a precautionary measure, all equipment used for chemical, radioactivity, and biological purposes must be clearly identified with appropriate labels, signs, or other conspicuous identification. ***High voltage electrical equipment must be labeled accordingly.***

Removal of Warnings: Radioactive and Biohazard Warning Labels can only be removed after appropriate decontamination or sterilization procedures have rendered them safe for further usage.

Multipurpose Warnings: The Safety Officer provides the necessary safety information concerning approved laboratory signage based on regulatory requirements. These signs have been or will be placed on the door(s) of the respective laboratory with the appropriate hazard warning information.

I. Electrical Equipment

Recommended procedures:

1. Inspect electrical equipment periodically for frayed cords, faulty control switches, and thermostats.
2. Do not try to repair any equipment yourself, contact the Building Manager immediately.
3. Never by-pass the ground or safety devices on a piece of electrical equipment.

J. Fire Safety

Fire safety is a precaution applicable to all personnel. At the Institute there are three basic elements to the Fire Safety Program.

Prevention: The ability to identify potential fire risk and eliminate them. Some of the procedures to eliminate fire risks are:

1. Practice Good Housekeeping
   a. Do not allow trash to accumulate
   b. Use the proper trash receptacles
   c. Keep combustibles to a minimum in your work area
   d. Keep flammable liquids properly stored
   e. Keep corridors, aisles and doors free of clutter to assure safe passage in the event of an emergency.
   f. Smoking is not permitted on any Institute properties.

2. Practice Electrical Safety
   a. Do not overload outlets
   b. Do not use extension cords in place of permanent wiring.
   c. Do not use damaged equipment.
   d. Do not store combustible or flammable material near electrical appliances that produce heat.

Detection: Even with a good program of prevention in place, the possibility of fire exists. In the event of fire remember

RACE **R** = rescue people in immediate danger.
A = alarm (activate a manual pull station and call the emergency operator).
C = contain the fire by closing the door to the room.
E = evacuate patients, visitors, students, and employees to safe areas.

Extinguish: In accordance with TUCA's Fire Safety Program, all employees are trained in the proper procedures regarding fire safety.

Types of Fire Extinguishers		
Class	**Material**	**Extinguisher**
Class A	Wood, Cloth, Paper	Water or dry chemical or Halon
Class B	Greases, Gasoline, Oils	CO_2 or dry chemical or Halon
Class C	Electrical Devices	Chemical or Halon
Class D	Combustible Metals	Special Technique

When Using the Extinguisher

Exercise good judgment when deciding to extinguish fire!

You must determine to "fight or flight."

PASS

P = pull the pin**.**
A = aim the extinguisher at the base of the fire.
S = squeeze the handle while holding the extinguisher upright.
S = sweep the nozzle from left to right to extinguish the fire.

Remember*: Do Not let the fire get between you and the exit!*

Accordingly, all personnel upon employment should know where the nearest:
a. location of the fire alarm is and how to use it.
b. fire extinguisher is and how to use it.
c. fire blanket is and how to use it.
d. evacuation route is.

If the fire alarm sounds – leave the building by the nearest (or designated) fire exit. Close all doors behind you (on the basis of last person out). Use stairways. Do not use elevators.

K. Visitors (Unauthorized Personnel)

Unauthorized personnel are prohibited from entering the laboratories and animal facilities. Individuals under 18 years of age, immunosuppressed persons, and pregnant visitors are not allowed to enter the laboratories of the Institute. As is the case for all personnel and visitors in a research laboratory, the Principal Investigator is responsible for training, assigning appropriate tasks and monitoring for safety practices.

L. Waste Disposal

Waste disposal depends on the nature of the material. Hazardous chemicals and flammable solvents are collected in special containers and disposed of by licensed Environmental Services. Biological and infectious waste (bleached or autoclaved) is collected by TUCA contracted licensed Environmental Services (call x8-5239 for information).

M. General Personnel Protection

1. Hallways and laboratory exits must never be blocked with equipment, file cabinets, and other laboratory supplies.
2. Safety showers and eye wash stations are located in designated areas of the facility; find the one nearest your laboratory. Safety showers and eye wash stations should be free from obstructions and hazards.
3. All volatile substances should be used in fume hoods and stored in explosion-proof solvent storage cabinets. Keep all flames away from volatile solvents.
4. Special gloves and safety glasses should be part of your laboratory equipment if the nature of your work includes danger of spills, breakage and explosive materials.
5. Ear protection should be used when working with high frequency sonic cell disrupters and homogenizers. Sonicators should be operated in a closed cabinet.
6. Glass jugs – preferably, safety-approved bottle carriers should be used instead of glass jugs for transporting liquids. When glass jugs are used, the jug should not be carried by the handle. Instead, one hand should support the base of the jug.

N. Pregnancy Protection

The pregnant woman and her fetus are uniquely susceptible to the effects of ionizing radiation, toxic chemicals, and infectious agents present in the laboratory. The time of greatest risk is the first 8 to 12 weeks of pregnancy, during which the woman may not know she is pregnant. The following precautions should be taken:

1. If you are pregnant, it is at your discretion to declare your pregnancy to the Institute's Radiation Safety Office and Employee Health Service. If you choose to declare your pregnancy, the radiation exposure limits will be reduced by a factor of ten (10).

2. Avoid using known teratogens (embryotoxins) if at all possible. Commonly used laboratory teratogens include formamide, organmercurials, lead compounds and anesthetic agents.

3. Discuss your work with your physician to determine what additional precautions should be followed. If your duties require you to work with infectious agents, consider all possible consequences to yourself and your child.

O. Emergencies/First Aid

First Aid Kits in laboratories and offices are for minor injuries. In the case of a serious injury or illness, call for emergency action immediately at 9-911 and Security at x8-5804.

P. Explosion-Proof Refrigerators & Freezers

Special explosion-proof freezers are located in the common-use rooms throughout the Institute for storage of flammable solvents. Under no circumstances, should flammable solvents be in normal (non explosion-proof) refrigerators or freezers.

Q. Absorbent Paper

Plastic-backed absorbent paper on laboratory bench tops will help control spills only if it is placed plastic side down.

R. Disinfectants

Disinfectants commonly used are: (a) Spor-Klenz containing hydrogen peroxide; (b) Clorox and Alcide containing sodium hypochlorite; (c) iodine compounds; (d) phenolics; (e) ammonium compounds, and (f) 70% alcohol. It is recommended that disinfectants have an Environmental Protection Agency (EPA) Registration Number and be effective against tuberculosis. The investigator should examine the expiration date, and determine whether the disinfectant is corrosive (ie: Clorox).

S. Autoclave Operation ***(only authorized personnel are allowed to operate autoclaves)***

See Appendix C for the guidelines to follow when operating an autoclave. Careful adherence to these precautionary measures will increase operational efficiency and help prevent accidents.

T. Mailing Etiological Agents

Based on the CDC recommendations, the U. S. Postal Service has adopted regulations for the packaging and labeling of etiological agents. According to these rules, such substances may be mailed only if they are intended for medical or research purposes and if they are properly packaged to prevent leaks or spills. All shipments and mailings are to be processed through the Shipping Department utilizing certified packers and labels. Specific restrictions and special permit requirements are mandated by federal guidelines, referred to as The Final Rule (*See Section VI – G & H for explanation of definitions and details for packaging, labeling, and mailing etiological elements.)*

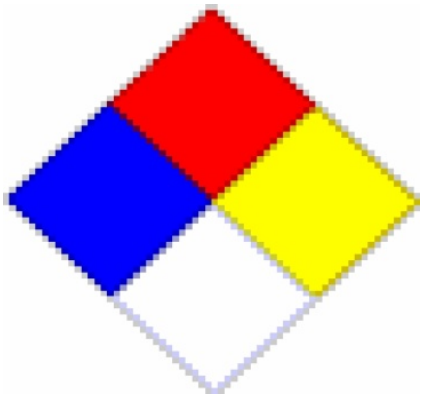
U. Vaccination

TUCA, in keeping with Section 6(b) of the OSHA Act of 1970, 29 U.S.C. 655, will offer hepatitis B vaccine at no cost to all employees at risk for HBV infection. The program can be scheduled through the Employee Health Clinic. The Institute will also offer all employees who may be at risk for other infectious agents (e.g., rabies, tetanus, and booster for chickenpox, measles, mumps, and rubella) a complete vaccination series free of charge through the Employee Health Clinic. The need for these specific vaccinations is determined by the supervisor in conjunction with the medical consultant and they are scheduled as needed.

Individuals requiring immunization for any infectious agents must sign a release form indicating their comprehension of the need for immunization and their agreement to be or not to be immunized.
Immunization requirements should be determined in conjunction with the Infection Control Plan which indicates who is at risk.

SECTION V – WARNING SIGNS

A. Hazardous Material Information System



Identify Hazard

Health – Blue	Flammability - Red	Reactivity – Yellow	Specific – White
4-Deadly	4-Flash point <73F	4-May detonate	*Oxy – Oxidizer*
3-Extreme Danger	3-Flash point <100F	3-Shock & Heat May Detonate	*Acid – Acid* *Alk – Alkali*
2-Hazardous	2-Flash point <200F	2-Violent Chemical Change	*Cor – Corrosive*
1-Slightly Hazardous	1-Flash point >200F	1-Unstable at Elev. Temp	~~*W*~~ *Use no water*
0-Normal Material	0-Will not burn	0-Stable	*Rad – Radiation*

Note: Each diamond represents a warning symbol based on the particular hazard classification.

WARNING SYMBOLS

B. Biohazard Warning Symbol

C. RadiationWarning Symbol

D. Laboratory Waste Disposal

Laboratory Material	Infectious and Sharps Waste (Red Bag, Cardboard Box, or Sharps Container)	Chemotherapeutic Waste (Yellow Bag)
Ampules	Yes	Yes, if chemotherapeutic waste
Animal Wastes	Yes, if exposed to zoonotic infection or human pathogens	Yes, if chemotherapeutic waste
Broken Glass	Yes	
Chemotherapeutic Agents (antineoplastics)	No	Yes, and designated w/a label for chemo
Chromatography Columns	Yes	
Cloning and sequencing equipment	Yes	
Cotton Tips Swabs (wooden)	Yes (*)	
Cover slips	Yes	
Culture Dishes	Yes	Yes, if chemotherapeutic waste
Culture Flasks	Yes	Yes, if chemotherapeutic waste
Culture tubes & tops, plastic	Yes	Yes, if chemotherapeutic waste
Electrophoresis plates	Yes	Yes, if chemotherapeutic waste
Gauze	Yes (*)	Yes, if chemotherapeutic waste
Rods (glass or hard plastic)	Yes	
Scalpel Blades	Yes	
Slides	Yes	
Specimen Containers, hard plastic or glass	Yes	

Specimen Containers, soft plastic	Yes	
Syringes/needles	Yes	
Test Tubes	Yes	
Tubing (glass)	Yes	
Cautery, hand held	Yes	
Patient Treatment Items	Yes (*)	Yes
Sharps Boxes, full	Yes (*)	Yes, if chemotherapeutic waste
Rods (glass or hard plastic)	Yes	
Scalpel Blades	Yes	
Slides	Yes	
Specimen Containers, hard plastic or glass	Yes	
Specimen Containers, soft plastic		
	Yes	
Syringes/needles		
	Yes	
Test Tubes		
	Yes	
Tubing (glass)		
	Yes	
Cautery, hand held		
	Yes	
Patient Treatment Items		
	Yes (*)	Yes
Sharps Boxes, full		
	Yes (*)	Yes, if chemotherapeutic waste

(*) If contaminated with blood/body fluids/infectious agents

SECTION VI – BIOLOGICAL SAFETY

A. Introduction

Almost any form of biological research involves the use of some potentially hazardous biological materials. A successful program to ensure biological safety and environmental control in the laboratory depends on careful observance of regulatory laws and meticulous attention to safe laboratory practices. The term "containment" is used in describing safe methods for managing infectious agents in the laboratory environment where they are being handled or maintained. The purpose of containment is to reduce or eliminate exposure of laboratory personnel, other persons and the outside environment to potentially hazardous agents. Emphasis should be placed on the use of containment equipment to protect laboratory personnel. In this regard, having each laboratory worker dedicated to maintaining good safety practices is the most important element in a safety program.

B. Risk Management

1. Risk Groups

Agents are classified into four Risk Groups (RGs) according to their relative pathogenicity for healthy adult humans by the following criteria:

a) Risk Group1 (RG1) (low individual and community risk). Any biological agent that is unlikely to cause disease in healthy workers or animals.

b) Risk Group 2 (RG2) (moderate individual risk, low community risk). Any pathogen that can cause human disease but, under normal circumstances, is unlikely to be a serious hazard to laboratory workers, the community, livestock or the environment.

c) Laboratory exposure rarely causes infection leading to serious disease; effective treatment and preventive measures are available, and the risk of spread is limited.

d) Risk Group 3 (RG3) (high individual risk, low community risk). Any pathogen that usually causes serious human disease or can result in serious economic consequences but does not ordinarily spread by casual contact from one individual to another, or that causes diseases treatable by antimicrobial or antiparasitic agents.

e) Risk Group 4 (RG4) (high individual risk, high community risk). Any pathogen that usually produces very serious human disease, often untreatable, and may be readily transmitted from one individual to another, or from animal to human or vice-versa, directly or indirectly, or by casual contact.

2. Criteria for Risk Groups

The classification of agents (See Appendix A) is based on the potential effect of a biological agent on a healthy adult and does not account for instances in which an individual may have

increased susceptibility to such agents. Such instances would include pre-existing diseases, medications, compromised immunity, pregnancy or breast feeding (which may increase exposure of infants to some agents). Personnel may need periodic medical surveillance to ascertain fitness to perform certain activities; they may also need to be offered prophylactic vaccines and boosters. Employee Health Services can be consulted as needed.

3. Comprehensive Risk Assessment

In deciding on the appropriate containment for an experiment, the initial risk assessment from the Classification of Human Etiologic Agents on the Basis of Hazard should be followed by a thorough consideration of the agent itself and how it is to be manipulated. Factors to be considered in determining the level of containment include agent factors such as: virulence, pathogenicity, infectious dose, environmental stability, route of spread, communicability, operations, quantity, availability of vaccine or treatment, and gene product effects such as toxicity, physiological activity, and allergenicity. Any strain that is known to be more hazardous than the parent (wild-type) strain should be considered for handling at a higher containment level. Certain attenuated strains or strains that have demonstrated irreversibly loss of known virulence factors may qualify for a reduction of the containment level compared to the Risk Group assigned to the parent strain. Biological risk assessment is a subjective process requiring consideration of many hazardous characteristics of agents and procedures, with judgments based often on incomplete information. The following 5 step approach gives structure to the risk assessment process:

1) Identify agent hazards and perform an initial assessment of risk.
2) Identify laboratory procedure hazards.
3) Make a final determination of the appropriate biosafety level and select additional precautions indicated by the risk assessment.
4) Evaluate the proficiencies of staff regarding safe practices and the integrity of safety equipment.
5) Review the risk assessment with a biosafety professional, subject matter expert, and IBC.

4. Biosafety Levels

Four BSLs are described, which consist of combinations of laboratory practices and techniques, safety equipment, and laboratory facilities. Each combination is specifically appropriate for the operations performed, the documented or suspected routes of transmission of the infectious agents, and the laboratory function or activity. The BSLs described should be differentiated from Risk Groups, as described in the NIH Guidelines and the World Health Organization Laboratory Biosafety Manual. Risk groups are the result of a classification of microbiological agents based on their association with, and resulting severity of, diseases in humans. The risk group of an agent should be one factor, to be considered in association with mode of transmission, procedural protocols, experience of staff, and other factors in determining the BSL in which the work will be conducted.

a) Biosafety Level 1

Suitable for work involving well characterized agents not known to cause disease in healthy adult humans, and of minimal potential hazard to laboratory personnel and the environment. The laboratory is not necessarily separated from the general traffic patterns in the building. Work is generally conducted on open bench tops using standard microbiological practices. Special containment equipment or facility design is not required nor generally used. Laboratory personnel have specific training in the procedures conducted in the laboratory and are supervised by a scientist with general training in microbiology or a related field.

b) Biosafety Level 2

Similar to Biosafety Level 1 and is suitable for work involving agents of moderate potential hazard to personnel and the environment. It differs in that (1) laboratory personnel have specific training in handling pathogenic agents and are directed by competent scientists, (2) access to the laboratory is limited when work is being conducted, (3) extreme precautions are taken with contaminated sharp items, and (4) certain procedures in which infectious aerosols or splashes may be created are conducted in biological safety cabinets or other physical containment equipment.

c) Biosafety Level 3

Applicable to clinical, diagnostic, teaching, research or production facilities in which work is done with indigenous or exotic agents which may cause serious or potentially lethal disease as a result of exposure by the inhalation route. Laboratory personnel have specific training in handling pathogenic and potentially lethal agents, and are supervised by competent scientists who are experienced in working with these agents.
All procedures involving the manipulation of infectious materials are conducted within biological safety cabinets by personnel wearing appropriate personal protective equipment. The laboratory has special engineering and design features. Access to the laboratory is strictly controlled by the Facility Manager or Principal Investigator. The facility is either in a separate building or in a controlled area within a building, which is completely isolated from all other areas of the building. A specific facility operations manual is prepared and adopted.

d) Biosafety Level 4

Required for work with dangerous and exotic agents which pose a high individual risk of aerosol-transmitted laboratory infections and life- threatening disease. Agents with a close or identical antigenic relationship to Biosafety Level 4 agents are handled at this level until sufficient data are obtained either to confirm continued work at this level, or to work with them at a lower level. Members of the laboratory staff have specific and thorough training in handling extremely hazardous infectious agents; and they understand the primary and secondary containment functions of the standard and special practices, the containment equipment, and the laboratory design characteristics. They are supervised by competent scientists who are trained and experienced in working with these agents.

a) Summary of Recommended Biosafety Levels for Infectious Agents

Biosafety Level	Agents	Practices	Safety Equipment	Facilities
1	Not known to cause disease in healthy Adults	Standard Microbiological Practices	None Required	Open bench top Sink required
2	Associated with human disease, hazard = auto-inoculation, ingestion, mucous membrane exposure	BSL1 practice plus: * Limited access; * Biohazard warning signs; * "Sharps" precautions; * Biosafety manual defining any needed waste decontamination or medical surveillance policies	Primary Barriers: Class I or II BSCs or other physical containment devices used for all manipulations of agents that cause splashes or aerosols of infectious materials; Personal Protective Equipment (PPEs); laboratory coat, gloves, face protection as needed.	BSL1 Plus: Autoclave available
3	Indigenous or exotic agents with potential for aerosol transmission; disease may have serious or lethal consequences.	BSL2 practice plus: * Controlled access; * Decontamination of all waste; * Decontamination of all lab clothing before laundering; * Baseline serum	Primary Barriers: Class 1 or II or BSCs or other physical containment devices used for all manipulations of agents that cause splashes or aerosols of infectious materials; Personal Protective Equipment(PPE); laboratory coats, gloves, respiratory protection as needed.	BSL2 plus: * Physical separation from corridors; * Self-closing double door access; * Exhausted air not recirculated; * Negative airflow into the laboratory

4	Dangerous/exotic agents which pose high risk of life threatening disease, aerosol transmitted lab infections, or related agents with unknown risk of Transmission	BSL3 practices plus: * Clothing change before entering; * Shower on exit; * All materials decontaminated on exit from facility	Primary Barriers: All procedures conducted in Class III BSCs in combination with full-body, air-supplied, positive pressure personnel suit.	BSL3 plus: * Separate building or isolated zone; * Dedicated supply/exhaust, vacuum, and decon systems; * Other requirements outlined in text.

C. Biological Safety Cabinets

The biological safety cabinet is the principal device used to provide containment of infectious splashes or aerosols generated by many microbiological procedures. This is an important function because most laboratory techniques are known to produce inadvertent aerosols that can be readily inhaled by laboratory personnel. Certain cabinets can also protect the experiment from airborne contamination. The selection of a biological safety cabinet is based on the potential hazard of the agent used in the experiment, the potential of the laboratory technique to produce aerosols, and the need to protect the experiment from airborne contamination. Three types of biological safety cabinets (Class I, II, and III) are used in the microbiological laboratories. Biological safety cabinets are designed to:

a. Protect personnel from harmful agents inside the cabinet (Personnel Protection). All three classes are effective.
b. Protect the work, product, experiment or procedure performed in the cabinet from contaminants in the laboratory environment or from cross-contamination inside the cabinet (Product Protection). Only Class II and III are effective.
c. Protect the environment from contaminants in the cabinet (Environmental Protection). All three classes are effective.

The NIH Guidelines for Recombinant DNA (See Appendix A) require that either the Class I or Class II Cabinet be used as the primary containment equipment when the BSL-2 or BSL-3 level of physical containment is specified. The Class III cabinet is required at the BSL-4 level of physical containment. The description, capabilities, and limitations of these cabinets follows:

Comparison of Biological Safety Cabinets

Type	Face Velocity (lfpm)	Airflow Patterns	Nonvolatile Toxic Chemicals and Radionuclides	Volatile Toxic Chemicals and Radionuclides	Biosafety Level(s)
Class I* open front	75	In at front, out rear and top through HEPA	Yes	Yes (1)	1-3
Class II A 1	75	70% recirculated through HEPA; 30% balance can be exhausted through HEPA back into the room or to the outside through a thimble unit	Yes	No	1-3
Class II A 2	100	Same as II A, but plena under negative pressure to room and exhaust air is ducted	Yes	No	1-3
Class II B1	100	30% recirculated through HEPA; exhaust via HEPA and hard ducted	Yes	Yes (minute amounts (2)	1-3
Class II B2	100	No recirculation, total exhaust via HEPA and hard ducted	Yes	Yes (small amounts)	1-3
Class III	NA	Supply air inlets and exhaust through 2 HEPA filters	Yes	Yes (small amounts)	4

(1) Installation may require a special duct to the outside, an in-line charcoal filter, and a spark proof (explosion proof) motor and other electrical components in the cabinet.
Discharge of a Class I cabinet in to a room should not occur if volatile chemicals are used.
(2) In no circumstances should the chemical concentration approach the lower explosion limits of the compound.
www.cdc.gov/od/ohs/biosfty/bsc/table2.htm

* Glove panels may be added and will increase face velocity to 150 lfpm, gloves may be added with an inlet air pressure release that will allow work with chemicals/radionuclides.

Note: The cabinets must be tested whenever they are installed, moved or whenever maintenance is performed, otherwise, they are tested once a year. Biosafety cabinets cannot be serviced or moved without approval from the Principal Investigator or the Biosafety Officer when necessary. Furthermore, mechanics must have a written order to work on any biosafety cabinet.

D. Recommended Safety Procedures

1. All centrifuges should be self-locking and investigators should use only unbreakable tubes. Before centrifuging tubes, motor and walls should be checked for damage.

2. Vacuum Lines, Filters, and Traps
   When the building vacuum line or when portable vacuum pumps are used, suitable traps or filters (such as Millipore or Gelman Vacushield filters) should be interposed to ensure that pathogens do not enter the central system. Vacuum flasks should contain disinfectants such as Clorox with a final concentration of 20%.

3. Freezers and Refrigerators
   Freezers and refrigerators should be cleaned-out periodically. All infectious or toxic material stored in refrigerators or freezers should be properly labeled. Do not place flammable solvents (i.e., ether) in normal refrigerators – use explosion-proof refrigerators and freezers.

4. Pipetting by mouth is forbidden. The only exception is mouse embryo transfer in the transgenic facility.

5. Use of Biosafety Cabinets Recommended procedures:
   a) Wipe down the work surface of the biosafety cabinet with a disinfectant (SporKlenz or Alcide followed by 20% ethanol). If the biosafety cabinet was turned off overnight, allow five (5) minutes of running time before starting your work.
   b) Assemble your materials and equipment BEFORE working in the biosafety cabinet.
   c) Minimize room activity, especially near the biosafety cabinet. Never walk behind someone working at a cabinet.
   d) Employ aseptic technique as you would on the bench-top. Separate clean from dirty items.
   e) Clean-up promptly and thoroughly when you are finished. Wipe down the work surface with disinfectant.
   f) Decontaminate any supplies that were used inside the biosafety cabinet.

6. Equipment
   All non-autoclaved equipment should be treated with disinfectant immediately after use. Disinfectants do not work instantaneously, but must be given several minutes to work before rinsing off.

7. Aerosol
   Sonification, blending, or any procedure that produces an infectious aerosol should be avoided. If it is necessary to perform these procedures, they must be carried out in a biological safety cabinet. Special precautions such as protective clothing and breathing devices should be used. Glass containers should not be used because of potential breakage. All instruments must be sterilized or disinfected after use.

8. Experimental Work with Infectious Agents
   Insure that all virulent fluid cultures or viable powdered infectious materials are transported, and stored in easily handled, non-breakable, sealed, leak-proof containers. Water baths used to inactivate or incubate infectious materials should contain a disinfectant.

9. Human Material with reference to bloodborne pathogens

   Investigators use body fluids and tissues for their experimental work. ***All materials should be treated as potentially infectious and handled as biohazards, using Standard Precautions.***

E. Disposal of Contaminated Materials

Paper and Disposable Plastics

All waste paper and plastic materials contaminated with potentially hazardous biological materials must be placed in red biohazard bags for disposal. They are to be sealed with tape, and disposed of through the red bag waste system. High risk material (BSL-3) should be autoclaved prior to disposal. Disposal of radioactive waste is described in the Radiation Safety Manual.

Recommended procedures:

a. Things To Be Returned:

All laboratory glassware
All bottles, except those used to contain toxic chemicals
Caps, stoppers, etc.
Pipette cans
Culture tube racks
Petri dish cans

b. Things NOT To be returned:

Chemicals
Radioactive materials
Animals or Animals parts
Plastic disposals

c. Methods to be followed:

Dirty glassware should be placed in plastic trays only after the trays have been lined with an autoclavable bag. Do not put dirty glassware in an unlined tray. Potentially dangerous items such as Pasteur pipettes, hypodermic needles, syringes, etc., are not to be returned for glassware washing.

d. Used Syringes, Needles and Pasteur Pipettes

Only syringes of the Luer-Lok type should be used with infectious materials. Used syringes, needles and Pasteur pipettes must be placed in an approved sharps container to be collected by TUCA contracted licensed Environmental Service.

F. Spill of Potential Biohazardous Materials

If an accident occurs involving the possible spread of potentially dangerous biologicals (virus, etc.), immediate steps must be taken to decontaminate the area. The amounts of material and hazards involved will determine the appropriate action.

1. Small Spills (To Not Exceed 50 ml)

For a small amount of liquid (not exceeding 50 ml with little or no virulence), use a paper towel

to absorb the spill, apply disinfectant (Clorox) to the area, let stand for a minimum of 10 minutes, and wipe-up. Rather than pour the disinfectant directly to the spill area and risk splashing, it is better to allow the disinfectant to flow onto the spill. Be sure to:

a. Use Standard Precautions when handling potentially biologically hazardous materials
b. Use double gloves.
c. Do not let the spill dry. A dried spill will allow contaminated dust to form and spread.
d. Dispose of absorbed materials into a biohazard bag.

2. Large Spills

For a large volume spill of virulent material:

a. Warn others
b. Leave the room.
c. Wash hands and any exposed body area.
d. Post a notice on the door to warn others not to enter the room.
e. Contact the Biosafety Officer with the exact location of the room and the nature of the spill.

Each researcher must realize that in the event of an overt accident, research materials such as tissue cultures, media, and animals within biological safety cabinets may be lost to the experiment.

G. Interstate Shipment of Infectious Agents

1. The following are the requirements for transportation of etiologic agents in interstate traffic recommended by the Department of Transportation and other Federal Government agencies.
49 CFR Part 171-178
Federal Register, Vol. 45, No. 141-Monday, July 21, 1980
Part 72-Interstate shipment of Etiologic Agents 1
Centers for Disease Control and Prevention
Office of Health and Safety Biosafety Branch
(Date Last Revised: March 9, 1995)

2. "Biological Substance, Category B" means any human or animal material including, but not limited to, excreta, secreta, blood and its components, tissue, and tissue fluids being shipped for purposes of diagnosis.

3. "Infectious Substance" means a viable micro-organism or its toxin which causes, or may cause, human disease. They are those micro-organisms that cause disease in humans and include bacteria, bacterial toxins, viruses, fungi, rickettsia, protozoans, and parasites. These disease causing micro-organisms may also be referred to as infectious agents or infectious substances. The materials such as body fluids and tissues that contain them are referred to as infectious materials. Organisms such as mosquitoes, that may transmit infectious diseases to other humans, are called vectors.

4. "Interstate Traffic" means the movement of any conveyance or the transportation of persons or property, including any portion of such movement or transportation which is entirely

5. within a state or possession: (a) from a point of origin in any state or possession to a point of destination in any other state or possession, or (b) between a point of origin and a point of destination in the same state or possession, but through any other state, possession, or contiguous foreign country. No person may knowingly transport or cause to be transported in interstate traffic, directly or indirectly, any material including, but not limited to, diagnostic specimens and biological products, if such person reasonably believes it may contain an etiologic agent. The exception being that such material is packaged to withstand leakage of contents, shocks, pressure changes, and other conditions incident to ordinary handling in transportation.

6. Volume not exceeding 50 ml.

Material should be placed in a securely closed, watertight and/or sift proof container (primary container (test tube, vial, etc.) which shall be enclosed in a second, durable watertight container (secondary container)). Several primary containers may be enclosed in a single secondary container, if the total volume of all the primary containers does not exceed 50 ml. The space at the top, bottom, and sides between the primary and secondary containers shall contain sufficient non-particulate absorbent material (e.g. paper towel) to absorb the entire contents of the primary container(s) in case of breakage or leakage. Each set of primary and secondary containers shall then be enclosed in an outer shipping container constructed of corrugated fiberboard, cardboard, wood, or other material of equivalent strength. Multiple primary containers should be wrapped individually so as not to touch.

7. Volume greater than 50 ml.

Packaging of material in volumes of 50 ml or more shall comply with requirements specified in #5 of this section. In addition, a shock absorbent material, in volume at least equal to that of the absorbent material between the primary and secondary containers, shall be placed at the top, bottom, and sides between the secondary container and the outer shipping container. Single primary containers shall not contain more than 1,000 ml of material. However, two or more primary containers whose combined volumes do not exceed 1,000 ml may be placed in a single, secondary container. The maximum amount of etiologic agent which may be enclosed within a single outer shipping container shall not exceed 4,000 ml.

8. Dry Ice

If dry ice is used as a refrigerant, it must be placed outside the secondary container(s). If dry ice is used between the second container and the outer shipping container, the shock absorbent material shall be placed so that the secondary container does not become loose inside the outer shipping container as the dry ice sublimates. The shipping container must allow for release of carbon dioxide gas.

9. Identification

The outer shipping container of all materials containing etiologic agents transported in interstate traffic must bear a label described below:

> The color of material on which the label is printed must be white, symbol red, and the printing in red or white.
>
> The label must be a rectangle measuring 51 millimeters (mm) (12 inches) by 10.25 mm (4 inches) long.

The red symbol measuring 38 mm (1-1/2 inches) in diameter must be centered in a white square measuring 51 mm (2 inches) on each side.

Type size of the letters of the label shall be as follows:

Etiologic agents – 10 pt.
Biomedical material – 14 pt.
In case of damage or leakage – 10 pt.
Notify Director CDC, Atlanta,Georgia – 8pt.
(404) 633-5313 – 10 pt.

e. An itemized list of contents between secondary and outer packaging.
f. Outer package must be of sufficient size to bear all necessary labels and possess strength for its capacity.
g. Completed packages must pass drop test.

10. Damaged Packages

The carrier shall promptly, upon discovery of evidence of leakage, or any other damage to packages, isolate the package and notify the Director for the Centers for Disease Control and Prevention, 1600 Clifton Road, N.E., Atlanta, Georgia 30333, by telephone (404) 633-5313. The carrier shall also notify the sender.

11. Registered Mail or Equivalent System

Transportation of the following etiologic agents shall be by registered mail, or an equivalent system, which requires or provides for sending notification of receipt to the sender immediately upon delivery:

Coccidioides Immitis
Ebola Virus
Francisella (Pasteurella) Tularensis
Hemorrhagic Fever agents including, but not limited to, Crimean
Hemorrhagic Fever (Congo), Junin, Machupo viruses, and Korean
Hemorrhagic Fever Viruses.
Herpesvirus Simiae (B virus)
Histoplasma Capsulatum
Lassa Virus
Marburg Virus
Pseudomonas Mallei
Pseudomonas Pseudomallei
Tick-borne Encephalitis Virus complex including, but not limited to
Russian Spring-Summer Encephalitis, Kyasanur Forest Disease, Omsk
Hemorrhagic Fever and Central European Encephalitis Viruses.
Variola Major and Variola Minor and White Pox Virus.
Yersinia (Pasteurella) Pestis.

12. Notice of Delivery, Failure to Receive

When notice of delivery of materials known to contain etiologic agents is not received by the sender within five days following the anticipated delivery of the package, the sender shall notify the Director for the Centers for Disease Control and Prevention, 1600 Clifton Road, N.E., Atlanta, Georgia 30333 by telephone (404) 633-5313

13. Requirements; Variations

The Director of CDC may approve variations from the requirements of this section if, upon review and evaluation, it is found that such variation(s) provide protection at least equivalent to that provided by compliance with the requirements specified in this section, and such findings are made a matter of official record.

H. Importation Permits for Etiologic Agents

Centers for Disease, Control and Prevention Etiologic Agent Import Permit Program

1. General

If you question whether your situation requires an importation permit, the safe alternative is to obtain and complete the application from the CDC's Department of Biosafety.

2. Importation Permits

Many etiologic agents, infectious materials or vectors containing infectious agents are imported from foreign locations into the United States for domestic use and study. Packages containing etiologic agents originating in these foreign locations must have an Importation Permit issued by the United States Public Health Service. Importation Permits are issued only to the importer, who must be located in the United States. The Importation Permit, with the proper packaging and labeling, will expedite clearance of the package of infectious materials through the United States Public Health Service Division of Quarantine and released by the U. S. Customs Department. The importer bears responsibility for assuring that the personnel for the foreign shipper pack and label the infectious materials according to USPHS Regulations. Transfers of previously imported material within the U.S. also require a permit for the same reason. Shipping labels containing the universal biohazard symbol, the address of the importer, the permit numbers, and the expiration date, are also issued to the importer with the permit. The importer must send the labels and one or more copies of the permit to the shipper. A label must be secured to each package and a copy of the permit should also be attached to the package. The permit and labels inform the U. S. Customs Service and the U. S. Division of Quarantine Personnel of the package contents.

3. Federal Regulations

The importation of etiologic agents is governed by the following federal regulation:

USPHS 42 CFR – Part 71 foreign Quarantine. Part 71.54 Etiologic agents, hosts, and vectors.

a. A person may not import into the United States, nor distribute after importation, any etiologic agent or any arthropod or other animal host or vector of human disease, or any exotic living arthropod or other animal capable of being a host or vector of human disease unless accompanied by a permit issued by the Director.

b. Any import coming within the provisions of this section will not be released from custody prior to receipt by the Port Director of the U. S. Customs Service of a permit issued by the Director of the CDC.

c. Letter of Authorization

After review of an "Application to Import an Etiological Agent" the issuing officer may issue a "Letter of Authorization" rather than an Importation Permit. The Letter of Authorization is issued for materials that are judged to be non-infectious, but which might be construed to be infectious by the U. S. Customs Inspection's personnel. Letters of Authorization may be issued for items such as formalin fixed tissues, sterile cell cultures, clinical materials such as human blood, serum, plasma, urine, cerebrospinal fluid, and other tissues or materials of human origin when there is no evidence or indication that such materials contain an infectious agent. A copy of a Letter of Authorization should be attached to the package, and furnished to the courier or importation broker. Letters of Authorization are in effect for two years, and do not require a shipping label to be issued by this office.

d. Packaging Requirements

Infectious materials imported into this country must be packaged to withstand leakage of contents and labeled as specified in the following federal regulations:
USPHS 42 CFR Part 72 – Interstate Shipment of Etiologic Agents
DOT 49 CFR Part 173 – Transportation of Etiologic Agents
For international shipments, the International Air Transport Association (IATA) Dangerous Goods Regulations should be consulted.

e. Other Permits

United States department of Agriculture (USDA) Animal and Plant Health Inspection Service (APHIS) Permits are required for infectious agents of livestock and biological materials containing animal materials, particularly livestock.

Tissue (cell) culture techniques customarily use bovine material as a stimulant for cell growth. Tissue culture materials and suspensions of cell culture grown viruses or other etiologic agents containing growth stimulants of bovine or other livestock origin are, therefore, controlled by the USDA due to the potential risk of the introduction of exotic animal diseases into the United States. Further information may be obtained by calling the USDA/APHIS at (301) 734-3277.

United States Department of Interior (USDI) Permits are required for certain live animals and all live bats. (Call (800) 358-2104 for further information).

f. Exports of Infectious Materials

The export of infectious material may require a license from the Department of Commerce. Call (202) 482-0896 for further information.

Centers for Disease Control and Prevention Office of Health and Safety
Biosafety Branch
1600 Clifton Road – MS F-05 Atlanta, Georgia 30333
Phone (404) 639-3235 Fax (404) 639-229

I. Final Rule: Additional Requirements for Facilities Transferring or Receiving Select Agents

This section is a summary of the ***Final Rule***, as stated in the Federal Register 42 CFR Part 71 On June 10, 1996, the CDC, the Department HHS issued a Notice of Proposed Rulemaking (NPRM) to implement Section 511 of Public Law 104-132, "The Antiterrorism and Effective Death Penalty Act of 1996," which requires the Secretary of HHS to regulate the transfer of select agents. Current regulations specify requirements for the packaging, labeling, and transporting of select agents shipped through interstate commerce. This Final Rule places additional shipping and handling requirements on facilities that transfer or receive select agents.

J. Select Agents:

Viruses:

1. Crimean-Congo Haemorrhagic Fever Virus
2. Eastern Equine Encephalitis Virus
3. Ebola Virus
4. Equine Morbillivirus
5. Lassa Fever Virus
6. Marburg Virus
7. Rift Valley Fever Virus
8. South American Haemorrhagic Fever Virus (Junin, Machupo, Sabia, Flexal, Guanarito).
9. Tick-borne Encephalitis Complex Viruses
10. Variola Major Virus (Smallpox Virus)
11. Venezuelan Equine Encephalitis Virus
12. Viruses causing Hantavirus Pulmonary Syndrome
13. Yellow Fever Virus

Rickettsia

1. *Coxiella Burnetii*
2. *Rickettsia Prowazekii*
3. *Rickettsia Rickettsii*

Fungi

Coccidioides Immitis

Toxins

1. Abrin
2. Aflatoxins
3. Botulinum Toxins
4. *Clostridium Perfringens* Epsilon Toxin
5. Conotoxins
6. Diacetoxyscirpenol
7. Ricin

8. Saxitoxin
9. Shigatoxin
10. Staphylococcal Enterotoxins
11. Tetrodotoxin
12. T-2 Toxin

K. Research Involving Recombinant DNA: (See Appendix A)

Experiments Using Risk Group 2, Risk Group 3, or Restricted Agents as Host Vector Systems
Experiments involving the introduction of recombinant DNA into Risk Group 2 Agents will usually be conducted at Biosafety Level (BSL-2) containment. Experiments with such agents will usually be conducted with whole animals at BSL-2 or BSL-2N (animals) containment. Experiments involving the introduction of recombinant DNA into Risk Group 3 Agents will usually be conducted at BSL-3 containment. Experiments with such agents will usually be conducted with whole animals at BSL-3 or BSL-3N containment.

Experiments in which DNA from Risk Group 2, Risk Group 3, or Restricted Agents is cloned into Non-Pathogenic Prokaryotic or Lower Eukaryotic Host-Vector System.
Experiments in which DNA from Risk Group 2 or Risk Group 3 Agents are transferred into non-pathogenic prokaryotes or lower eukaryotes may be performed under BSL-2 containment. The Institute's Biosafety Committee may approve the specific lowering of containment for particular experiments to BSL-1. Experiments involving the formation of recombinant DNA for certain gene encoding for molecules toxic for vertebrates require NIHOBA approval. Experiments involving the cloning of toxin molecules with LD_{50} (of less than 100 nanograms per kilogram body weight) shall be conducted under NIH specified conditions. Containment conditions for experiments in which DNA from select agents is transferred into non-pathogenic prokaryotes or lower eukaryotes shall be determined by NIHOBA following a case-by-case review.

Recombinant DNA or RNA molecules derived from any source except for greater than two-thirds of eukaryotic viral genome may be transferred to any non-human vertebrate or any invertebrate organism and propagated under conditions of physical containment comparable to BSL-1 or BSL-1N and appropriate to the organism under study. Animals that contain sequences from viral vectors, which do not lead to transmissible infection either directly or indirectly as a result of complementation or recombination in animals, may be propagated under conditions of physical containment comparable to BSL-1 or BSL-1N and appropriate to the organism under study. It is important that the investigator demonstrates that the fraction of the viral genome being utilized does not lead to productive infection.

Experiments that Require Institutional Biosafety Committee Notice Simultaneous with Initiation

Recombinant DNA molecules containing no more than two-thirds of the genome of any eukaryotic virus (all viruses from a single family being considered identical) may be propagated and maintained in cells in tissue culture using BSL-1 containment. For such experiments, it must be demonstrated that the cells lack a helper virus for the specific families of defective viruses being used. The DNA vector may contain fragments of the genome of viruses from more than one family, but each fragment shall be less than two-thirds of a genome. Experiments in which all

components derived from non-pathogenic prokaryotes and non-pathogenic lower eukaryotes may be conducted at BSL-1 containment.

Biosafety Considerations for Research with Lentiviral Vectors

A comprehensive risk assessment and determination of containment for research with lentiviral vectors should consider the nature of the vector system, transgene insert, and type of manipulations involved. For many experiments, either BSL-2 or enhanced BSL-2 will be appropriate. For more information visit the OBA website at: www4.od.nih.gov/oba/rac/Guidance/LentiVirus_Containment/index.htm Questions about the guidance may be directed to Marina O'Reilly, Ph.D., Biotechnology Program Advisor, NIH OBA, at 301-496-9838.

Exempt Experiments

The following recombinant DNA molecules are exempt from the NIH Guidelines, and registration with the Institute's Biosafety Committee is not required.

1. Those that are not performed in organisms or viruses
2. Those that consist entirely of DNA segments from a single non-chromosomal source, though one or more of the segments may be a synthetic equivalent.
3. Those that consist entirely of DNA from a prokaryotic host including its indigenous plasmids or viruses when propagated only in the host (or a closely related strain of the same species).
4. Those that consist entirely of DNA from an eukaryotic host including its chloroplasts, mitochondria, or plasmids (but excluding viruses) when propagated only in that host (or a closely related strain of the same species).
5. Those that consist entirely of DNA segments from different species that exchange DNA by known physiological processes, though one or more of the segments may be a synthetic equivalent. A list of such exchangers will be prepared and periodically revised by the NIH Director with advice of the RAC after appropriate notice and opportunity for public comment.
6. Those that do not present a significant risk to health or the environment as determined by the NIH Director, with the advice of the RAC, and following appropriate notice and opportunity for public comment.

For Your Information

Summary of the changes in NIH Guidelines for Recombinant DNA in relation to transgenic rodents:

1. Purchase or transfer of transgenic animals (from commercial or non-commercial sources) is exempt from the NIH guidelines and approval of the Institute's Biosafety Committee, provided the animals and experiments can be carried out at BSL-1 containment.
2. Generating new transgenic animals requires notification to the Institute's Biosafety Committee at the initiation of the experiment, provided the animals and experiments can be carried out at BSL-1 containment. Generating transgenics using DNA sequences from Risk Group 2 or 3 Agents will require prior approval by the Institute's Biosafety Committee for the appropriate containment level.

SECTION VII – CHEMICAL SAFETY

CHEMICAL SAFETY

A. Introduction

All employees need to understand the hazards of chemicals in the workplace. Individually, everyone must take the time to read the labels of the chemicals which they work with, learn how to recognize the hazards (both physical and chemical) which the product poses, how to avoid the hazards, and what to do in case of an "exposure".

B. General Information

1. OSHA's Hazard Communication Standard

The major points pertaining to chemical safety contained in the Hazard Communication Standard (29 CFR 1910.1200), "Right To Know" are:

a. Hazard Identification – Identify all hazardous chemicals in the workplace.
b. Labeling – Identify the hazards on all chemical containers used in the laboratory.
c. MSDS – Material Safety Data Sheets must be available for all chemicals identified in OSHA's Hazard Communication Standard. Laboratories are required to keep all of their own Material Safety Data Sheets on file in the laboratory.
d. Employee Training and Information – All employees must be informed that the Right-To-Know Law exists, its scope and their rights as workers exposed to hazardous chemicals. The employer must provide any specialized training necessary to protect the laboratory worker(s) from existing chemical hazards. (See Appendix A).

The Institute's Biosafety Committee, as well as OSHA, requires that each research laboratory maintain a chemical inventory and MSDS for each chemical it possesses. An updated Chemical Inventory will be forwarded to the Safety Office on a biennial basis or as new products are being utilized in the research laboratory.

The Committee encourages safe laboratory practices at all times. The Committee encourages the PI and laboratory personnel to pay special attention when handling regulated carcinogen; highly hazardous chemicals, toxics, reactives; highly flammable chemicals (rated #4 on flammability chart); peroxidizable; and shock sensitive substances. The Safety Office is available to assist any laboratory in answering questions and addressing concerns about the safe handling, appropriate storage and disposal of chemicals.

2. Flammable Storage Cabinets

All work with noxious, poisonous, dangerous, concentrated acids, flammable liquids and gaseous substances should be conducted in a fume hood. Flammables should be stored in labeled and grounded flammable storage cabinets. Under no circumstances should flammables be stored in non-explosion proof refrigerators. When working with flammables, all flames in the laboratory should be extinguished prior to their use.

3. Storage Areas and Transport

Laboratory bench tops and aisles should not be used as storage areas; they should be cleared upon completion of each experiment. Basket carriers should be used when transporting glass containers holding concentrated acids, etc. To avoid accidents or mistaken identity, chemicals should be stored in easy to reach (below eye-level) shelving.

4. Acid Cleaning Solutions

Acid cleaning solutions are very dangerous, especially to the eyes. Safety goggles and rubber gloves should be used when working with these solutions. Cleaning fluid should be kept in polyvinyl chloride containers, rather than glass.

5. Chemical Spills
    a. Small spills (i.e., < 50 ml involving no chemical hazard to personnel):
    Will be handled by laboratory personnel, utilizing spill kits located in common areas.
    b. Large spills (i.e., > 50 ml)
    Security is to be contacted immediately at extension x8-5804, or Biosafety Officer at x8-5239 for prompt clean-up and proper disposal.

6. Mercury and Mercury Spills

Care should be taken with mercury and compounds containing mercury. Disposable gloves and respiratory protection should be used when working with compounds and solutions containing mercury. Mercury spills are to be cleaned-up by individuals who have received training using mercury spill kits. Mercury waste is to be labeled for laboratory pak disposal or recycling. In case of a spill, contact the Biosafety Oficer at x8-5239. If the spill occurs after normal working hours, contact the Security Department at x8-5804.

7. Waste Disposal

Small and diluted quantities of inorganic acids or alkali (pH levels between 3 and 11) can be disposed of by slowly pouring into a stream of faucet water. The drain should then be thoroughly flushed with a large quantity of water. Organic compounds and heavy metals should not be poured down the drain. Agencies such as the local water department, the Department of Environmental Conservation (DEC), and the Environmental Protection Agency (EPA) have jurisdiction in matters concerning the disposal of chemicals in the wastewater treatment system. Any concerns regarding what compounds can be disposed via the drain should be directed to the Department of Occupational & Environmental Safety.

Waste materials can be disposed of by completing the Request for Waste Disposal Form on TUCA's Webpage and forwarding it to the BSO. Special arrangements will then be made for collecting and disposing of waste material. See TUCA Waste Disposal Guide for more information on the handling, storage and disposal of waste materials.

C. Fume Hoods

Fume hoods are an important engineering control designed to protect laboratory workers from potential chemical exposures. Always work with hazardous chemicals in a fume hood, do not use hazardous chemicals on the bench top.

1. Follow the guidelines outlined in the Safe Fume Hood Use Guide.
2. Always work 6 inches back from the sash of the fume hood to achieve the best capture of chemical vapors and fumes.
3. Always work with the fume hood sash as low as possible and keep your fume hood sash closed when you are not working in it (this offers better protection against splashes and explosions).
4. Always report any malfunctioning fume hoods to the Building Coordinator immediately to have the hood repaired. If the hood is not working properly, let other people in the lab know this by hanging up a sign on the fume hood.

D. Carcinogens

Precautionary measures, similar to those pertaining to radioactive compounds and biohazardous materials are required when working with or storing carcinogens.
Laboratory work with chemical carcinogens will be permitted in posted areas only. Safeguards as described in "*The NIH Guidelines for the Laboratory Use of Chemical Carcinogens"* provide recommended procedures for minimizing exposure of laboratory personnel to chemical substances that pose a carcinogenic risk.
Recommended Procedures:
1. Use disposable plasticware.
2. Minimal protective clothing should include a laboratory coat, rubber gloves, and respirator. Some carcinogens may need to be handled in a chemical hood.
3. Storage and work areas are marked with the appropriate signs. A locked storage box should be considered (with a key kept in another location).
4. Transport of carcinogens to other laboratories or the animal facility should be done in unbreakable transport containers lined with adequate absorbent.

E. Selecting The Appropriate Gloves

www.ansellpro.com/download/Ansell_7thEditionChemicalResistanceGuide.pdf
www.pacifica.com/NitrileGlovesChemicalResistance-BarrierGuide.pdf (See Appendix A)

SECTION VIII – RADIATION SAFETY

A. Introduction

The Radiation Safety Program at the Institute is administered by the Radiation Safety Office under the direction of the Biosafety Committee. The use of radioactive materials is regulated by various agencies including the U.S. Nuclear Regulatory Commission and the California State Department of Public Health (CDPH). CDPH issues a license which specifically authorizes the use of various radionuclides. The license has conditions and limitations to which the Institute employees must adhere.

Investigators who desire to use radioactive materials in their laboratories need to apply for authorization from the Biosafety Committee. The Committee reviews the training and experience

of the Investigator with regard to the proposed use of radioactive materials to determine that the work is within the conditions and limitations of the Institute's state license and that the facilities and equipment are adequate to perform the work safely, and the procedures are conducted to minimize the risk of exposure and contamination. Investigators that have been approved to use radioactive materials in their laboratories have the responsibility to adhere to any conditions of approval and compliance with the Radiation Safety Program.

Contact the Radiation Officer to request personal monitoring (film badges and ring TL dosimeters); to report accidents, incidents, or spills; to request services, or if there are any questions regarding proper procedures.

For emergencies after normal business hours, contact Security at x8-5804.

The Radiation Safety Officer provides a variety of services including:

1. Monitoring radiation exposures, including surveys, personal monitoring, and bioassays.
2. Investigating spills and incidents involving radioactive materials.
3. Investigating over-exposures.
4. Conducting an ALARA Program to keep radiation exposures As Low As Reasonably Achievable.
5. Maintaining an inventory of all radioactive materials at the Institute.
6. Disposing of radioactive waste.
7. Surveying packages of radioactive materials upon receipt.
8. Transporting radioactive materials in compliance with the U.S. Department of Transportation requirements.
9. Auditing and inspecting laboratories for compliance with the Radiation Safety Program.
10. Conducting independent surveys of laboratories.
11. Training radiation workers through short course, in-services, and meetings.
12. Approving users and uses of radioactive materials.
13. Maintaining requisite records.

B. Radioisotope Licensing

The Radiation Safety Office of the Institute is responsible for issuing licenses to investigators, monitoring the procedures for safety and health involving all individual users, monitoring isotope records, and disposal of all radioactive materials. Please consult with the Radiation Safety Office for information concerning issuance of new, modified, or the transfer of licenses.

C. Radiation Warning Signs

Radiation warning signs must be placed on the doors of laboratories using radioactive materials. Laboratory equipment such as refrigerators, sinks, centrifuges, containers, etc. used for laboratory procedures involving radioactive materials should be marked with radiation hazard warning labels.

D. Radiation Work Areas

Work areas for the exclusive use of radioactive materials should be established in the laboratory. Absorbent paper with non-porous backing and/or spill trays should cover the work surfaces to contain spills.

E. General Radiation Safety Procedures

All licensees of radioactive materials are required to return a copy of their completed inventory/disposal sheets to the Radiation Safety Office in accordance with the instructions contained in the Institute's Radiation Safety Manual and the Institute's Polices and Procedures.

1. Eating, drinking, chewing gum, application of cosmetics, manipulation of contact lenses is NOT permitted.
2. Do not store foodstuffs for human consumption in laboratory.
3. Wear laboratory coat.
4. Wear disposable gloves and change gloves often.
5. Use drip trays where practical.
6. Use plastic backed absorbent paper on work area.
7. Label radioactive work area with radioactive warning tape.
8. Seal containers of radioactive material when vortexing, centrifuging, and incubating.
9. Use a secondary trap flask in series with collection flask for vacuum aspiration.
10. Wear radiation monitoring badge(s) if assigned.
11. Only wear the radiation monitoring badge assigned to you.
12. Post the following notices on laboratory doors in the laboratory

NOTICE TO EMPLOYEES
RADIATION SAFETY PROCEDURES
CAUTION – RADIOACTIVE MATERIALS

13. Label the laboratory equipment with a "Caution Radioactive Material" tape.
14. Maintain record of receipt, use, and disposal of radioactive materials.
15. Monitor hands, shoes, and clothing after each use.
16. Wash hands after using radioactive materials and when leaving the work area. Survey yourself (hands, body, and feet) and work area after each use of radioactive materials.
17. Dispose of waste according to the guidelines in the Radiation Safety Manual and the Institute's Policies & Procedures.
18. Follow the procedures for receiving radioactive material packages.
19. Use appropriate shielding.
20. Prevent unauthorized access to radioactive materials by challenging unauthorized individuals, locking radioactive materials, or locking the laboratory when no one is physically present.
21. Follow the approved protocol and any conditions of authorization.

F. Emergency Instructions

Specific instructions for handling emergencies should be posted in each laboratory. The most current version of the Emergency Procedures can be obtained from the Radiation Safety Manual.

G. Package Receipt

Most packages containing radioactive materials must be surveyed for contamination upon receipt. Regulations require formal procedures for safely opening packages containing radionuclides. The initial package check-in is performed by the Radiation Safety Officer, or his designee. Refer to the Radiation Safety Manual for details.

H. Radioactive Waste Disposal

Refer to the Radiation Safety Manual for radioactive waste disposal procedures.

I. Pregnant Workers

State and Federal Regulations limit the radiation dose to the embryo/fetus of an occupationally exposed declared pregnant woman to 0.5 rem (500 millirem) for the entire gestation period. A declared pregnant woman is defined in these regulations as "a woman who has voluntarily informed the licensee, in writing, of her pregnancy and the estimated date of conception."

1. The dose limit only considers the occupational dose. Any radiation exposure received as a patient from medical diagnosis or treatment, and natural background radiation are not considered.
2. A pregnant woman may seek recommendations from the Radiation Safety Officer to reduce radiation exposure to her embryo/fetus without declaring her pregnancy.
3. The woman must provide the Declaration of Pregnancy in writing to impose the more restrictive limit.
4. The Declaration of Pregnancy is strictly voluntary.

In effect, a pregnant woman has the choice of declaring her pregnancy, thereby imposing a dose limit to her embryo/fetus. To comply with the more restrictive radiation dose limits, the Institute may require the use of additional protective equipment (e.g., additional shielding, lead aprons), increased monitoring (e.g. extra film badges, pocket dosimeters), or re-assign work duties. Note that most activities involving exposure to radiation at the Institute results in annual radiation exposures less than 500 millirem.

J. Policy and Procedures

To comply with this regulation, the Institute has implemented the following policy and procedures:

1. The pregnant woman who wishes to impose radiation dose limits for her embryo/fetus must provide a written declaration to the Radiation Safety Officer that includes the estimated date of conception.
2. A Declaration of Pregnancy is strictly voluntary.
3. A pregnant woman who plans to declare her pregnancy is encouraged to do so promptly upon discovering her pregnancy so that the appropriate precautions can be taken early in the gestation period.

4. The Declaration of Pregnancy will be kept confidential. The Declaration of Pregnancy will only be disclosed to Institute employees with legitimate need-to-know (e.g. immediate supervisor).
5. Any woman may request additional information on the risks associated with radiation exposure to the embryo/fetus from the Radiation Safety Officer.
6. The declared pregnant worker will notify the Radiation Safety Officer of the end of her pregnancy so that the special precautions can be terminated.
7. The radiation dose limit to the embryo/fetus of a declared pregnant woman is 0.5 rem (500 millirem). The radiation dose limit applies only to occupational exposure of the declared pregnant woman. It does not apply to radiation exposure from medical diagnosis or treatment.
8. Restrictions may be imposed to prevent radiation exposures from exceeding 500 millirem during the gestation. These restrictions may include a temporary change in work assignments, the use of additional protective equipment, and increased monitoring.
9. If the embryo/fetus radiation exposure has exceeded 450 millirem before the pregnancy is declared, a dose limit of 50 millirem will be in effect for the remainder of the pregnancy.

K. Minor Spills (involving no radiation hazard to personnel)

1. Notify all other persons in the room or area that a spill has occurred.
2. Prevent the spread of contamination by covering the spill with absorbent paper.
3. Decontaminate the area using paper towels or absorbent pads. Clean towards the center of the spill. Place all waste into a plastic bag and dispose of it as radioactive waste. Disposable gloves, laboratory coats and, if appropriate, shoe covers should be worn. Cleansing agents may be used after initial decontamination attempt.
4. Survey the area and all contaminated and potentially contaminated individuals with a G-M Survey Meter. Survey for removable contamination using wipe samples.
5. Report the incident to the Radiation Safety Officer.

L. Major Spills (involving potential radiation hazard to personnel, involving personal contamination, involving actual or potential uptake or radioactive materials, or which threatens to restrict the use of the facility)

1. Clear the area: notify all persons not involved with or near the spill to vacate the room.
2. Prevent spread of contamination. Cover the spill with absorbent paper. Do NOT attempt to clean it up. Assemble all potentially contaminated personnel near the room entrance.
3. Close the room: prevent entry into the room.
4. Call for help: immediately contact the Radiation Safety Office.
5. Decontaminate personnel: survey personnel for contamination. Contaminated clothing should be removed and stored for evaluation by Radiation Safety. Contaminated skin should be flushed thoroughly and then washed with mild soap and lukewarm water.

M. Fires

1. **R**escue: persons in immediate danger.
2. **A**larm: activate manual pull station and call Security at x8-5804 with the fire location.

3. **C**ontain the fire by closing the door to the room.
4. **E**vacuate the area – do not attempt to extinguish the fire unless:
   a. The fire presents an immediate risk of injury to you or someone else in the area.
   b. The fire is very small in size, easily extinguished, and you have had fire extinguisher training.

Do NOT attempt to extinguish the fire if radioactive materials are directly involved. Evacuate the area; contact Radiation Safety, and notify the firefighters that radioactive materials are involved.

During normal working hours, Contact Radiation Safety Officer
After normal working hours, call 9-911 and Security at x8-5804.

Section IX – Laboratory Animals

A. Care and Use of Laboratory Animals

Special attention must be given to the humane treatment of all laboratory animals in accordance with the Animal Welfare Act of 1996 as amended, the Public Health Service Policy on the Humane Care and Use of Laboratory Animals and the policies of the Institute.

The Institute's Attending Veterinarian and IACUC establishes procedures to ensure the use of animals that are free of disease prejudicial to the proposed experiments, and free from carriers of disease or vectors such as ectoparasites, which endanger other experimental animals or personnel.

Animal care technicians are well trained in the basic fundamentals of laboratory animal care. Appropriate training materials are available from a number of animal care associations or commercial organizations. Animal care technicians, scientists, or others routinely exposed to infected animals, potentially contaminated equipment, and animal waste must participate in preventative medical training and the medical surveillance programs of the Institute.

1. Care and Handling of Infected Animals

There are 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 of activities involving infected laboratory animals. The ABSLs describe animal facilities and practices applicable to work with animals infected with agents assigned to the appropriate Biosafety Levels. http://www.cdc.gov/od/ohs/biosfty/bmbl4/bmbl4s4.htm

Comprehensive reviews indicate that animals infected with a wide range of etiological agents are capable of shedding infectious micro-organisms in the saliva, urine, or feces. In the absence of specific information to the contrary, all infected animals should be regarded as potential shedders.

Procedures appropriate for the handling of infected animals are given below:

a. Trained personnel carry out all the procedures including necropsies using certified Biological Safety Cabinets.
b. Necropsies of potentially infected animals must be carried out under the same conditions, and additional precautions should be taken according to the specified hazards.
c. Personal Protective Equipment (PPEs) – gowns, head covers, shoe covers and, if necessary, face masks are worn at all times while inside the animal facilities
d. In addition to the PPEs, when handling the animals, gloves should be worn at all times.
e. It is recommended that for necropsies, dedicated instruments and an appropriate board to position the animal be utilized. The board should be either disposable or made from a material that can be disinfected.
f. All of the supplies for sample collection including containers, swabs for cultures, slides, etc. should be prepared in advance.
g. Upon completion of each necropsy, all potentially biohazardous materials should be disposed of in the appropriate labeled receptacles.
h. The animal carcasses should be double bagged, appropriately labeled and placed in the freezer.
i. The cages should be placed in the labeled racks indicating that they should be autoclaved before taken to the cage wash area.
j. Instruments and other supplies should be disinfected and carefully cleaned following the procedures approved by the Institute's Biosafety Committee and then autoclaved or disposed.

2. General Guidelines that Apply to Animal Room Maintenance

a. Doors to animal rooms should be kept closed at all times, except for necessary entrances and exits.
b. Unauthorized persons should not be permitted to enter animal rooms.
c. A container of disinfectant is kept in each biohazard suite for disinfecting gloves and hands, and for general decontamination even though no infectious animals are present. Hands, floors, walls, and cage racks are washed with an approved disinfectant at the recommended strength as frequently as the supervisor directs.
d. Floor drains in animal rooms, as well as floor drains throughout the building, should be flooded with water or disinfectant periodically to prevent backup of sewer gases.
e. Animal bedding and other refuse on floors should not be washed down the floor drain because such refuse clogs the sewer lines.
f. An insect and rodent control program should be maintained in all animal rooms and in animal food storage areas.
g. Specific care should be taken to prevent live animals, especially mice, from finding their way into disposable trash.
h. Specific instructions involving the housing, care, and maintenance of laboratory animals are available from the following sources:
    1. Laboratory Safety Monograph, A Supplement to the NIH Guidelines for recombinant DNA Research, January 1979.
    2. Occupational Health and Safety in the Care and Use of Research Animals, 1997.
    3. Guide for the Care and Use of Laboratory Animals, 1996.
    4. Biosafety in Microbiological and Biomedical Laboratories, CDC/NIH 2007.

B. Cage Cleaning

Biohazard cages should only be handled by staff that is formally trained in biohazardous and zoonotic diseases.

1. Biohazardous animals must be contained in designated rooms with negative air flow and a sign posted on the room door with the following information:
   a. PI name and contact person's phone number.
   b. Protocol number.
   c. Identification of hazard.
2. Cages must be identified by the use of biohazard labels in front of cages.
3. All personnel manipulating cages must wear personnel protective equipment including:
   a. Gown
   b. Disposable gloves
   c. Shoe covers
   d. Head cover
   e. Surgical or N95 mask depending on the situation
4. All cage changing must be performed in an approved/certified biosafety cabinet.
5. Biosafety cabinets must be cleaned with the proper disinfectant before and after each use.
6. Animals are transferred from dirty cage to clean cage using forceps that are decontaminated between cages.
7. After cage changing, all cages (including bedding, food, and water devices) are placed on a designated rack on the fourth floor of MRC.
8. All biohazardous cages must be autoclaved prior to delivery to cage processing.
9. All scientists are required to comply with institute policy regarding cage cleaning.
10. All animal cages contaminated with chemicals must be cleaned in compliance with institute policy

C. Transportation of Research Animals

1. Research animals will only be transported in DLAR/IACUC approved vehicles. All animals will be transported in escape-proof filtered-top cages or shipping containers. These containers must provide adequate ventilation and can be autoclaved or disposed of to prevent the spread of pathogenic micro-organisms.
2. Care shall be exercised in handling enclosures used to transport live animals. They should not be tossed, dropped, tilted or stacked in a manner that may cause physical trauma or stress to the animals.
3. Temperature extremes should be avoided. Postponements are required when temperatures are below 45^{o}F or above 85^{o}F.
4. Biohazard labels will be placed on the front or top of the containers. Labels must state type of hazard, protocol number, PI name and phone number.
5. The transportation of animals that are to be housed at one location and moved to another need to be evaluated to assure that proper containment is used to minimize occupational exposure to persons involved with the move and to minimize environmental contamination.

6. Small laboratory animals that have been exposed to human pathogens or toxic/carcinogenic

substances, and are actively shedding the hazardous material, must be transported in closed systems.

D. Visitors (Unauthorized Personnel)

Unauthorized personnel are prohibited from entering the laboratories and animal facilities. Individuals under years of age, immunosuppressed persons, and pregnant visitors are forbidden to enter the laboratories of the Institute. As is the case for all personnel and visitors in a research laboratory, the Principal Investigator is responsible for training, assigning appropriate tasks and monitoring for safety practices. In addition, the Principal Investigator will decide who may be admitted to his/her laboratory in accordance with Federal and State Law, as well as TUCA's Policy and Procedures.

APPENDIX A

REFERENCES AVAILABLE

Centers for Disease Control (CDC)
Biosafety in Microbiological and Biomedical Laboratories (BMBL)
http://www.cdc.gov/OD/ohs/biosfty/bmbl4/bmbl4toc.htm

National Institute of Health (NIH) Guidelines for Research Involving Recombinant DNA Molecules (NIH Guidelines)
http://www4.od.nih.gov/oba/rac/guidelines/guidelines.html

Centers for Disease Control (CDC) Select Agent Program
http://www.cdc.gov/od/sap

Occupational Safety and Health Administration (OSHA) blood-borne pathogen (29 CFR 1910.1030) and
Needle-stick Prevention Standards
http://www.osha.gov/SLTC/bloodbornepathogens/standards.html

Centers for Disease Control (CDC) National Institute for Occupational Safety & Health (NIOSH)
http://www.cdc.gov/niosh/topics/chemical-safety

California Department of Public Health (CDPH)
http://www.cdph.ca.gov/Pages/DEFAULT.aspx

Selecting Gloves
www.ansellpro.com/download/Ansell_7thEditionChemicalResistanceGuide.pdf
http://www.pacifica.com/NitrileGlovesChemicalResistance-BarrierGuide.pdf

Office of Biosafety Administration
http://www4.od.nih.gov/oba/rac/Guidance/LentiVirus_Containment/index.htm

TUCA Safety Committee, Biosafety Committee
and Material Safety Data Sheets

APPENDIX B

INFECTIOUS AGENTS – RISK GROUPS

Bacteria
Viruses
Fungi
Parasites

http://www.absa.org/resriskgroup.html

APPENDIX C

BIOLOGICAL SAFETY

Autoclave Operation

WHAT TO DO	HOW TO DO IT	KEY POINTS WHY
Prepare for start-up	Remove plug screen from bottom of chamber and clean.	If plugged with debris, will interfere with free flow of steam
Arrange load	Place flat packs of supplies on edge. If several tiers, place alternate tiers crosswise.	To ensure adequate flow of steam.
Loading containers of liquid	Do not mix loads of liquids with other supplies.	
	Use only vented closures	Sealed bottles may explode.
	Use only type 1 borosilicate (Pyrex) glass bottles	Stress of temperature and pressure may rupture ordinary glass.
	Use sterilizer slow exhaust only	Fast exhaust causes rapid boiling within the bottles with loss of fluids. Do not place flammable chemicals, chemical which are unstable at high temperatures in the sterilizer.
	Moisten loads of cloth or fabric	Dry fabrics remove moisture from steam, causing superheating which chars the fabric.
Close autoclave door	Turn handle clockwise until arms are within rim of door – continue turning handle until snug. To sterilize all materials: Turn timer to desired exposure period.	Time required for sterilization varies with load.
	Turn selector to appropriate position: “slow exhaust” for liquids; “fast exhaust” for drying.	

APPENDIX D

IMPORTATION PERMITS

Importation Permits are issued by the Etiologic Agent Import Permit Program at the Centers for Disease Control and Prevention (CDC) following review of a completed application form. The regulation, application and instructions can be found at the CDC website http://www.cdc.gov/od/eaipp/ . Completed application forms may be returned to the CDC, Etiologic Agent Import Permit Program by Fax: 404-718-2093 or by mail to: Centers for Disease Control and Prevention, Etiologic Agent Import Permit Program, 1600 Clifton Road, N.E., Mailstop A-46, Atlanta, GA 30333.

Application for the Importation Permit should be made fifteen (15) working days in advance of the shipment date to allow time for processing, issuance, and delivery of the permit and shipping labels to the permittee.