Auburn University
Biological Safety Manual

August 2009

(Incorporating by Reference National Institutes of Health (NIH) Guidelines for Research Involving Recombinant DNA Materials and the Centers for Disease Control and Prevention/NIH Biosafety in Microbiological and Biomedical Laboratories (BMBL))

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Emergency Phone Numbers and Resources

Risk Management and Safety	(334) 844-4870
After Work Hours	911
City of Auburn Department of Public Safety	911

Other Important Numbers

Centers for Disease Control and Prevention	(404) 639-3534 or (800) 311-3435
State Veterinarian, Alabama	(334) 240-7255
Office of Biotechnology Activities, NIH	(301) 496-9838
AU Office of Animal Resources	(334) 844-5978
AU Office of Human Subjects Research	(334) 844-5966
AU Scientific Supply Store	(334) 844-6940

(Source of Auburn University approved autoclave bags)

Select Agent Program Contacts:

Responsible Official: Margaret Smith:	(334) 703-2359
Alternate Responsible Official: Shawn McNulty	(334) 740-9711

Web Resources

Risk Management and Safety	http://www.auburn.edu/administration/rms/
Centers for Disease Control and Prevention	http://www.cdc.gov/
National Institutes of Health	http://www.nih.gov/
National Institute for Occupational Safety and Health	http://www.cdc.gov/niosh/homepage.html

I. Preface

This manual, in conjunction with the latest edition of the Centers for Disease Control and Prevention (CDC)/National Institutes of Health (NIH) Biosafety in Microbiological and Biomedical Laboratories (BMBL), provides the requirements for safely working with biohazardous materials at Auburn University (AU). The Institutional Biosafety Committee (IBC) is responsible for the regulation of biohazardous materials on the Auburn University campus. Principal Investigators (PIs) using biohazardous materials within Auburn University facilities or by Auburn University faculty, staff, or students must register with the IBC using what is termed a Biological Use Authorization (BUA). The IBC may be contacted through the Office of the Vice President for Research at 844-5966. Specific responsibilities of PIs, the IBC, and other involved parties are included in Section VI. Many investigators may work with vertebrate animals and/or radioisotopes in conjunction with their work with some biohazardous agents. The Institutional Review Board for the Protection of Human Subjects in Research (IRB) reviews all human subject research conducted at the University or by Auburn University faculty, staff, or students. All vertebrate animal work conducted at Auburn University must be approved by the Institutional Animal Care and Use Committee (IACUC) which can be contacted at 844-5978. All work with radioisotopes conducted at Auburn University must be licensed through the Radiological Safety Committee which can be contacted at 844-4870.

The primary concerns when working with biohazardous materials is the safety of personnel working with the agent and prevention of the release of that agent. Traditionally, microorganisms have been classified by the biological containment they require. Four levels of containment have been defined and termed Biosafety Levels (BSLs or BLs 1-4). Similarly, organisms are assigned classes dependant on the biosafety level they require. For example, an organism requiring BL2 precautions is designated as a Class 2 agent or organism. The specific requirements of BLs are found in the BMBL, an integral component of the Biological Safety Manual. NIH has introduced the concept of four Risk Groups (RGs) by which agents are classified, based on their relative pathogenicity for healthy adult humans. In almost all instances, the RG correlates with the recommended containment level. Thus, an RG2 organism is usually handled using BL-2 containment. Under some circumstances, the containment level required may be raised or lowered as a result of a comprehensive risk assessment. Auburn University has adopted the concept of RGs as discussed in Section II of the manual.

The BUA is Auburn University's registration document for use with biohazardous agents and materials. The forms shall be completed as applicable to the research being conducted. BUAs are required for all instructional, research, and outreach projects involving potentially biohazardous microorganisms; etiologic agents; infectious agents; oncogenic viruses; human

tissue and blood borne pathogens; and in vitro construction or propagation of recombinant DNA molecules.

Standard Operating Procedures (SOPs) for BL 1-3 laboratory operations are included in Appendix A of this manual. Work with biohazardous agents or materials at AU facilities shall be performed in accordance with (IAW) these SOPs.

Waste management procedures are discussed in the *Auburn University Medical Waste Management Guide*. The State of Alabama has defined medical waste as sharps or any items displaying the biohazard symbol or wording to that effect. Therefore, even new red bags, unopened syringes and gas chromatography syringes are medical waste once discarded. In general, red bags or autoclave bags displaying the biohazard symbol or wording to that effect are not to be used at AU. Instead blue autoclave bags are available through the AU Scientific Supply Store. Blue autoclave bags may be discarded as normal trash after autoclaving.

Training requirements are summarized in Appendix E. These include a series of video-tapes on basic laboratory safety; lab and agent specific training provided by the PI; and medical waste and bloodborne pathogen training provided by RMS as necessary. These constitute the minimum training requirements. Contact RMS for assistance in determining further training needs.

The PI has primary responsibility for the safety of students, faculty, staff, visitors, and the environment with respect to their laboratory operations. Specific questions concerning this manual or the Auburn University Biological Safety Program can be directed to RMS at 844-4870. PIs should be aware of other safety programs applicable to their laboratory operations including the Laboratory Safety Manual and Chemical Waste Management Program. These references are available on-line at Risk Management and Safety's web page.

Biohazardous Materials Policy

Introduction

Biohazardous materials are materials of biological origin that could potentially cause harm to humans, animals, or plants. Examples include recombinant DNA; transgenic animals or plants; human, animal or plant pathogens; biological toxins (such as aflatoxin); human blood and other potentially infectious materials; and human or non-human primate cell cultures.

The Institutional Biosafety Committee (IBC) has responsibility for reviewing the biological safety programs at Auburn University and sets policies that comply with federal, state, and local regulations. The IBC members are appointed by the President.

The IBC reviews research projects to ensure compliance with National Institutes of Health (NIH) and the Centers for Disease Control and Prevention (CDC)/NIH *Biosafety in Microbiological and Biomedical Laboratories* guidelines. An institution receiving NIH funding for any of its recombinant DNA projects must follow NIH Guidelines in all of its recombinant DNA research projects.

Policy

Approval for Use

The Institutional Biosafety Committee (IBC) must approve and issue a Biological Use Authorization (BUA) for any teaching or research project that involves:

- Use of recombinant DNA, including transgenic animals or plants
- Use of human, animal, or plant pathogens not indigenous to Alabama or the region
- Use of biological toxins; administration of experimental biological products to animals
- Field releases of plant pests (not indigenous to Alabama) or genetically modified organisms.

It is Auburn University policy that no Risk Group 4 agents or any agent requiring the use of biosafety level 4 as defined by the NIH may be used or stored at AU. Further, AU prohibits work that requires BL3-P or BL4-P containment.

Non-Human Primates

Non-human primates are not allowed at Auburn University.

Training

Annual training is required for:

- Biosafety
- Laboratory Safety
- Bloodborne Pathogens (if applicable).
- All personnel working in BL-3 laboratories must attend an accredited BL-3 training course. Prior documented experience (less than 2 years prior) may be substituted for this requirement.

II. Definitions

Biosafety Levels

The essential elements of the four biosafety levels for activities involving infectious microorganisms and laboratory animals are described in this section. The levels are designated in ascending order, by degree of protection provided to personnel, the environment, and the community.

Biosafety Level 1-

is suitable for work involving well-characterized agents not known to consistently 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 neither 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 science.

Biosafety Level 2-

is similar to Biosafety Level 1 and is suitable for work involving agents of moderate potential hazard to personnel and the environment. It differs from BSL-1 in that (1) laboratory personnel have specific training in handling pathogenic agents and are directed by scientists competent with this level of research/investigation; (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.

Biosafety Level 3-

is 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 scientists competent and experienced in working with these agents. The Auburn University Institutional Biosafety Committee strictly limits and regulates the work with BL3 agents and the space required for these operations.

Biosafety Level 4 is required for work with dangerous and exotic agents that pose a high individual risk of life-threatening disease, aerosol transmission, or related agent with unknown risk of transmission. Agents with a close or identical antigenic relationship to agents requiring BSL-4 containment must be handled at this level until sufficient data are obtained either to confirm continued work at this level, or re-designate the level. Laboratory staff must have specific and thorough training in handling extremely hazardous infectious

agents. Laboratory staff must understand the primary and secondary containment functions of standard and special practices, containment equipment, and laboratory design characteristics. All laboratory staff and supervisors must be competent in handling agents and procedures requiring BSL-4 containment. Auburn University does not currently have a BSL 4 laboratory and work with BSL 4 agents is strictly prohibited.

Plant Biosafety Levels

There are four plant biosafety levels, designated Plant Biosafety Level 1 through 4 (BL1-P through BL4-P), for work with plant diseases in whole plants or transgenic plants. The levels designate combinations of practices, safety equipment and facilities for experiments on plants infected with agents that produce or may produce disease in cultivated crops or transgenic plants. In general, the biosafety level recommended for working with an infectious agent in vivo and in vitro are comparable. At this time, Auburn University policy does not allow work requiring BL3-P or BL4-P containment.

The use of any greenhouse for research with transgenic plants must be approved by the IBC and responsible greenhouse management personnel.

Plant Biosafety Levels are discussed in Appendix C.

Animal Biosafety Levels

All animal use in AU facilities will comply with IACUC and IBC requirements. Work at Animal Biosafety Levels 1 and 2 are permitted. Facilities do not currently exist for work at higher biocontainment levels.

Animal Biosafety Level 1 (ABSL1) is suitable for work involving well characterized agents that are not known to cause disease in healthy adult humans, and that are of minimal potential hazard to laboratory personnel and the environment.

Animal Biosafety Level 2 (ABSL2) involves practices for work with those agents associated with human disease. It addresses hazards from ingestion as well as from percutaneous and mucous membrane exposure. ABSL2 builds upon the practices, procedures, containment equipment, and facility requirements of ABSL1.

Risk Groups

Traditionally, microorganisms have been classified according to the biological containment they require. Four levels of containment have been defined and termed Biosafety Levels (BSLs or BLs 1-4). More recently, the NIH has introduced the concept of Risk Groups (RGs) based on their relative pathogenicity for healthy adult humans. The following table lists the four groups and the basis for classification.

Risk Group 1 (RG1)	Agents that are not associated with disease in healthy adult humans
Risk Group 2 (RG2)	Agents that are associated with human disease which is rarely serious and for which preventive or therapeutic interventions are often available
Risk Group 3 (RG3)	Agents that are associated with serious or lethal human disease for which preventive or therapeutic interventions may be available (high individual risk but low community risk)
Risk Group 4 (RG4)	Agents that are likely to cause serious or lethal human disease for which preventive or therapeutic interventions are not usually available (high individual risk and high community risk)

Biohazardous Material

Biohazardous materials are materials of biological origin that could potentially cause harm to humans, animals, or plants. Examples include recombinant DNA; transgenic animals or plants; human, animal or plant pathogens; biological toxins (such as aflatoxin); human blood and other potentially infectious materials; and human or non-human primate cell cultures.

Human Blood and Other Potentially Infectious Materials

Experimentation or manipulation of human blood or other materials of human origin, including, but not limited to: excreta, secreta, blood and its components, unfixed tissue, and tissue fluids, all of which may or may not contain an infectious agent, may place the worker at risk of exposure to bloodborne pathogens. Examples which could result in exposure would be clinical laboratories performing tests or analyses on human blood or other potentially infectious materials, research labs performing experiments and/or manipulations with human blood or unfixed tissues or organs. Any work with human blood or other materials of human origin may need to be approved by the Institutional Review Board (IRB.)

Infectious Agents and Materials

Infectious agents, pathogens or substances are defined as those substances containing viable microorganisms or their toxins which are known or suspected to cause disease in animals, plants or humans. Pathogens are classified as bacteria, fungi, rickettsia, viruses, parasites, oncogenic viruses, and prions. Any materials which come in contact with infectious agents or their byproducts must be handled and disposed of in the same manner appropriate for disposal of the infectious agents themselves.

Recombinant DNA Molecules and Products

Research involving experiments with rDNA materials includes, but is not limited to: commonly used host-vector systems such as E. coli; recombinant

DNA experiments using whole animals or plants; recombinant DNA or RNA experiments involving infectious animal or plant viruses; the production of transgenic animals; and the deliberate transfer of DNA or RNA into human subjects (requires IRB approval and approval from federal agencies, including NIH). Regardless of the cloning method utilized, precautions must be taken to assure that the systems neither cause disease in the operator nor release recombinant molecules into the environment. For more information please see the NIH guidelines.

Miscellaneous Biohazardous Materials

These include materials not directly covered by the above definitions, such as; allergens, cultured animal cells and their potentially infectious agents, tissues from experimental animals (including animal dander), plant viruses, bacteria and fungi, toxins (bacterial, plant, etc.), and those as yet unnamed elements or agents which may produce disease. In regard to allergens, it is not the intent of Auburn University to require a BUA for research projects involving only allergens (i.e., projects involving allergens not listed as biohazardous agents or materials). However, it is important that PIs consider the hazard associated with allergens because manipulations of these allergens, or materials containing allergens, may result in human exposures. The control of allergens in any teaching, research or outreach project must be an important consideration in experimental/procedural design, because severe allergenic reactions can be life-threatening. Prior to initiation of work, personnel must be instructed on the appropriate use of safety devices and procedures used to minimize exposures.

Mixed Waste

Mixed wastes are potentially infectious materials contaminated with other types of hazardous materials, e.g., radioisotopes or toxic/carcinogenic compounds. Because waste disposal is controlled by more than one set of requirements and/or regulatory agencies, it is critical that provisions be made for proper management prior to the initiation of any research resulting in mixed waste. Mixed wastes may require special containers, labeling, storage, etc. Contact RMS (844-4805) prior to initiation of any research that might generate mixed waste.

III. Biosafety Guidelines

Biosafety Level 1

The following standard practices, safety equipment, and facility requirements apply to BSL-1:

A. Standard Microbiological Practices

1. All laboratory doors shall remain closed and locked unless personnel are present in the lab. In addition, laboratory doors must remain closed when agents are being manipulated in the laboratory.
2. Persons must wash their hands after working with potentially hazardous materials and before leaving the laboratory.
3. Eating, drinking, smoking, handling contact lenses, use of tobacco products, applying cosmetics, and storing food for human consumption is not permitted in laboratory areas. Food must be stored outside the laboratory area in cabinets or refrigerators designated and used for this purpose.
4. The laboratory supervisor must document that laboratory personnel demonstrate proficiency in standard and special microbiological practices before working with BSL-2 agents.
5. Mouth pipetting is prohibited; mechanical pipetting devices must be used.
6. Procedures for the safe handling of sharps, such as needles, scalpels, and pipettes must be developed and implemented at the laboratory level. Whenever practical, laboratory supervisors should adopt improved engineering and work practice controls that reduce risk of sharps injuries. Precautions, including those listed below, must always be taken with sharp items. These include:
    a. Careful management of needles and other sharps are of primary importance. Needles must not be bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated by hand before disposal.
    b. Used disposable needles and syringes must be carefully placed in conveniently located puncture-resistant containers used for sharps disposal.
    c. Non disposable sharps must be placed in a hard walled container for transport to a processing area for decontamination, preferably by autoclaving.
7. Broken glassware shall be handled according to the university's procedures.
8. Perform all procedures to minimize the creation of splashes and/or aerosols.
9. Decontaminate work surfaces after completion of work and after any spill or splash of potentially infectious material with appropriate disinfectant.
10. Decontaminate all cultures, stocks, and other potentially infectious materials before disposal using an effective method.
11. Materials to be decontaminated outside of the immediate laboratory must be double bagged, placed in a durable leak proof container and secured for transport.
12. All laboratories shall utilize the Door Sign Program available on the RMS website. Laboratories shall designate the appropriate biosafety level and after hours contact information on the sign. In addition, NO agent specific information shall be posted on this sign.
13. An effective integrated pest management program is required.

14. The laboratory supervisor must ensure that laboratory personnel receive appropriate training regarding their duties, the necessary precautions to prevent exposures, and exposure evaluation procedures. Personnel must receive annual updates or additional training when procedural or policy changes occur. All training must be documented.

B. Safety Equipment (Primary Barriers and Personal Protective Equipment)

1. Protective laboratory coats, gowns, or uniforms are recommended to prevent contamination of personal clothing.
2. Wear protective eyewear when conducting procedures that have the potential to create splashes of microorganisms or other hazardous materials. Persons who wear contact lenses in laboratories should also wear eye protection.
3. Gloves must be worn to protect hands from exposure to hazardous materials. Glove selection should be based on an appropriate risk assessment. Alternatives to latex gloves should be available. Wash hands prior to leaving the laboratory. In addition, BSL-1 workers should:
    a. Change gloves when contaminated, integrity has been compromised, or when otherwise necessary.
    b. Remove gloves and wash hands when work with hazardous materials has been completed and before leaving the laboratory.
    c. Do not wash or reuse disposable gloves. Dispose of used gloves with other contaminated laboratory waste. Hand washing protocols must be rigorously followed.

C. Laboratory Facilities (Secondary Barriers)

1. Laboratories must have a sink for hand washing.
2. The laboratory should be designed so that it can be easily cleaned. Carpets and rugs in laboratories are not appropriate.
3. Laboratory furniture must be capable of supporting anticipated loads and uses.
4. Spaces between benches, cabinets, and equipment should be accessible for cleaning.
5. Bench tops must be impervious to water and resistant to heat, organic solvents, acids, alkalis, and other chemicals.
6. Chairs used in laboratory work must be covered with a non-porous material that can be easily cleaned and decontaminated with appropriate disinfectant.
7. Laboratory windows that open to the exterior should be fitted with screens.

Biosafety Level 2

The following standard and special practices, safety equipment, and facility requirements apply to BSL-2:

A. Standard Microbiological Practices

1. All laboratory doors shall remain closed and locked unless personnel are present in the lab. In addition, laboratory doors must remain closed when agents are being manipulated in the laboratory.
2. Persons must wash their hands after working with potentially hazardous materials and before leaving the laboratory.
3. Eating, drinking, smoking, handling contact lenses, applying cosmetics, use of tobacco products, and storing food for human consumption must not be permitted in laboratory areas. Food must be stored outside the laboratory area in cabinets or refrigerators designated and used for this purpose.
4. Mouth pipetting is prohibited; mechanical pipetting devices must be used.
5. Procedures for the safe handling of sharps, such as needles, scalpels, and pipettes must be developed and implemented at the laboratory level. Whenever practical, laboratory supervisors should adopt improved engineering and work practice controls that reduce risk of sharps injuries. Precautions, including those listed below, must always be taken with sharp items. These include:
    a. Careful management of needles and other sharps are of primary importance. Needles must not be bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated by hand before disposal.
    b. Used disposable needles and syringes must be carefully placed in conveniently located, appropriately labeled, puncture-resistant containers used for sharps disposal.
    c. Non disposable sharps must be placed in a hard walled container for transport to a processing area for decontamination, preferably by autoclaving.
6. Broken glassware shall be handled according to the university's procedures.
7. Perform all procedures to minimize the creation of splashes and/or aerosols.
8. Decontaminate work surfaces after completion of work and after any spill or splash of potentially infectious material with appropriate disinfectant.
9. Decontaminate all cultures, stocks, and other potentially infectious materials before disposal using an effective method.
10. Materials to be decontaminated outside of the immediate laboratory must be placed in a labeled, durable, leak proof container and secured for transport.
11. Materials to be removed from the facility for decontamination must be packed in accordance with applicable local, state, and federal regulations.
12. A sign incorporating the universal biohazard symbol must be posted at the entrance to the laboratory when infectious agents are present. Posted information must include: the laboratory's biosafety level, the supervisor's name (or other responsible personnel), telephone number, and required procedures for entering and exiting the laboratory. Agent information should be posted in accordance with the institutional policy.
13. All laboratories shall utilize the Door Sign Program available on the RMS website. Laboratories shall designate the appropriate biosafety level and after hours contact

information on the sign. In addition, NO agent specific information shall be posted on this sign.

14. An effective integrated pest management program is required.
15. The laboratory supervisor must ensure that laboratory personnel receive appropriate training regarding their duties, the necessary precautions to prevent exposures, and exposure evaluation procedures. Personnel must receive annual updates or additional training when procedural or policy changes occur. All training must be documented.

B. Special Practices

1. All persons entering the laboratory must be advised of the potential hazards and meet specific entry/exit requirements.
2. Laboratory personnel must be provided medical surveillance and offered appropriate immunizations for agents handled or potentially present in the laboratory.
3. The biosafety manual must be available and accessible.
4. The laboratory supervisor must document that laboratory personnel demonstrate proficiency in standard and special microbiological practices before working with BSL-2 agents.
5. Potentially infectious materials must be placed in an appropriately labeled, durable, leak proof container during collection, handling, processing, storage, or transport within a facility.
6. Laboratory equipment should be routinely decontaminated, as well as, after spills, splashes, or other potential contamination.
    a. Spills involving infectious materials must be contained, decontaminated, and cleaned up by staff properly trained and equipped to work with infectious material.
    b. Equipment must be decontaminated before repair, maintenance, or removal from the laboratory.
7. Incidents that may result in exposure to infectious materials must be immediately evaluated and treated. Medical treatment shall follow the AU policy for On-the-Job Injuries as posted on the RMS website. All such incidents must also be reported to the laboratory supervisor.
8. Animals and plants not associated with the work being performed must not be permitted in the laboratory.
9. All procedures involving the manipulation of infectious materials that may generate an aerosol should be conducted within a biosafety cabinet (BSC) or other physical containment devices.

C. Safety Equipment (Primary Barriers and Personal Protective Equipment)

1. Properly maintained BSC, other appropriate personal protective equipment, or other physical containment devices must be used whenever:
    a. Procedures with a potential for creating infectious aerosols or splashes are conducted. These may include pipetting, centrifuging, grinding, blending, shaking, mixing, sonicating, opening containers of infectious materials, inoculating animals intranasally, and harvesting infected tissues from animals or eggs.

    b. High concentrations or large volumes of infectious agents are used. Such materials may be centrifuged in the open laboratory using sealed rotor heads or centrifuge safety cups.
2. Protective laboratory coats, gowns, smocks, or uniforms designated for laboratory use must be worn while working with hazardous materials. Remove protective clothing before leaving for non-laboratory areas (e.g., cafeteria, library, administrative offices). Dispose of protective clothing appropriately, or deposit it for laundering by the institution. It is recommended that laboratory clothing not be taken home.
3. Eye and face protection (goggles, mask, face shield or other splatter guard) is used for anticipated splashes or sprays of infectious or other hazardous materials when the microorganisms must be handled outside the BSC or containment device. Eye and face protection must be disposed of with other contaminated laboratory waste or decontaminated before reuse. Persons who wear contact lenses in laboratories should also wear eye protection.
4. Gloves must be worn to protect hands from exposure to hazardous materials. Glove usage shall be compatible with the materials used. Gloves must not be worn outside the laboratory. In addition, BSL-2 laboratory workers should:
    a. Change gloves when contaminated, integrity has been compromised, or when otherwise necessary. Wear two pairs of gloves when appropriate.
    b. Remove gloves and wash hands when work with hazardous materials has been completed and before leaving the laboratory.
    c. Do not wash or reuse disposable gloves. Dispose of used gloves with other contaminated laboratory waste. Hand washing protocols must be rigorously followed.
5. Eye and face protection should be used in rooms containing infected animals.

D. Laboratory Facilities (Secondary Barriers)

1. Laboratory doors should be self-closing and remain locked when laboratory personnel are not present and closed when biohazardous materials are being manipulated.
2. Laboratories must have a sink for hand washing. The sink may be manually, hands-free, or automatically operated.
3. Carpets and rugs in laboratories are not permitted.
4. Laboratory furniture must be capable of supporting anticipated loads and uses.
5. Spaces between benches, cabinets, and equipment should be accessible for cleaning.
    a. Bench tops must be impervious to water and resistant to heat, organic solvents, acids, alkalis, and other chemicals.
    b. Chairs used in laboratory work must be covered with a non-porous material that can be easily cleaned and decontaminated with appropriate disinfectant.
6. If a laboratory does have windows that open to the exterior, they must be fitted with screens.
7. Vacuum lines should be protected with High Efficiency Particulate Air (HEPA) filters, or their equivalent. Filters must be replaced as needed.
8. An eyewash station must be readily available.
9. A method for decontaminating all laboratory wastes should be available in the

10. Facility (e.g., autoclave, chemical disinfection, incineration, or other validated decontamination method).

Biosafety Level 3

The following standard and special safety practices, equipment, and facility requirements apply to BSL-3:

A. Standard Microbiological Practices

1. All BL-3 laboratories will have key card controlled access (or other secure method.) These laboratories will remain locked at all times.
2. Persons must wash their hands after working with potentially hazardous materials and before leaving the laboratory.
3. Eating, drinking, smoking, handling contact lenses, use of tobacco products, applying cosmetics, and storing food for human consumption must not be permitted in laboratory areas. Food must be stored outside the laboratory area in cabinets or refrigerators designated and used for this purpose.
4. Mouth pipetting is prohibited; mechanical pipetting devices must be used.
5. Procedures for the safe handling of sharps, such as needles, scalpels, and pipettes must be developed and implemented at the laboratory level. Whenever practical, laboratory supervisors should adopt improved engineering and work practice controls that reduce risk of sharps injuries. Precautions, including those listed below, must always be taken with sharp items. These include:
    a. Careful management of needles and other sharps are of primary importance. Needles must not be bent, sheared, broken, recapped, removed from disposable syringes, or otherwise manipulated by hand before disposal.
    b. Used disposable needles and syringes must be carefully placed in conveniently located, appropriately labeled, puncture-resistant containers used for sharps disposal.
    c. Non disposable sharps must be placed in a hard walled container for transport to a processing area for decontamination, preferably by autoclaving.
6. Broken glassware shall be handled according to the university's procedures.
7. Perform all procedures to minimize the creation of splashes and/or aerosols.
8. Decontaminate work surfaces after completion of work and after any spill or splash of potentially infectious material with appropriate disinfectant.
9. Decontaminate all cultures, stocks, and other potentially infectious materials before disposal using an effective method. A method for decontaminating all laboratory wastes will be available in the facility (e.g., autoclave, chemical disinfection, or other validated decontamination method).
10. All laboratories shall utilize the Door Sign Program available on the RMS website. Laboratories shall designate the appropriate biosafety level and after hours contact information on the sign. In addition, NO agent specific information shall be posted on this sign.
11. The laboratory supervisor must ensure that laboratory personnel receive appropriate training regarding their duties, the necessary precautions to prevent exposures, and exposure evaluation procedures. Personnel must receive annual updates or additional training when procedural or policy changes occur. All training must be documented.

B. Special Practices

1. All persons entering the laboratory must be advised of the potential hazards and meet specific entry/exit requirements. This must be documented and documentation maintained for 3 years.
2. Laboratory personnel must be provided medical surveillance and offered appropriate immunizations for agents handled or potentially present in the laboratory.
3. A laboratory-specific biosafety manual must be prepared and adopted as policy. The biosafety manual must be available and accessible.
4. The laboratory supervisor must ensure and document that laboratory personnel demonstrate proficiency in standard and special microbiological practices before working with BSL-3 agents.
5. Potentially infectious materials must be placed in a durable, leak proof container during collection, handling, processing, storage, or transport within a facility.
6. Laboratory equipment should be routinely decontaminated, as well as, after spills, splashes, or other potential contamination.
    a. Spills involving infectious materials must be contained, decontaminated, and cleaned up by staff properly trained and equipped to work with infectious material.
    b. Equipment must be decontaminated before repair, maintenance, or removal from the laboratory.
7. Incidents that may result in exposure to infectious materials must be immediately evaluated and treated. Medical treatment shall follow the AU policy for On-the-Job Injuries as posted on the RMS website. All such incidents must also be reported to the laboratory supervisor.
8. Animals and plants not associated with the work being performed are not permitted in the laboratory.
9. All procedures involving the manipulation of infectious materials must be conducted within a BSC, or other physical containment devices. No work with open vessels is conducted on the bench. When a procedure cannot be performed within a BSC, a combination of personal protective equipment and other containment devices, such as a centrifuge safety cup or sealed rotor, must be used.

C. Safety Equipment (Primary Barriers and Personal Protective Equipment)

1. All procedures involving the manipulation of infectious materials must be conducted within a BSC, or other physical containment devices.
2. Protective laboratory clothing with a solid-front such as tie-back or wraparound gowns, scrub suits, or coveralls are worn by workers when in the laboratory. Protective clothing is not worn outside of the laboratory. Reusable clothing is decontaminated with appropriate disinfectant before being laundered. Clothing is changed when contaminated.
3. Eye and face protection (goggles, mask, face shield or other splatter guard) is used for anticipated splashes or sprays of infectious or other hazardous materials. Eye and face protection must be disposed of with other contaminated laboratory waste or decontaminated before reuse. Persons who wear contact lenses in laboratories must also wear eye protection.

4. Gloves must be worn to protect hands from exposure to hazardous materials. Glove selection should be based on an appropriate risk assessment. Alternatives to latex gloves should be available. Gloves must not be worn outside the laboratory. In addition, BSL-3 laboratory workers should:
    a. Change gloves when contaminated, integrity has been compromised, or when otherwise necessary. Wear two pairs of gloves when appropriate.
    b. Remove gloves and wash hands when work with hazardous materials has been completed and before leaving the laboratory.
    c. Do not wash or reuse disposable gloves. Dispose of used gloves with other contaminated laboratory waste. Hand washing protocols must be rigorously followed.
5. Eye, face, and respiratory protection must be used in rooms containing infected animals. Users using respiratory protection must be medically fit and fit to the respirator prior to entering the area.

D. Laboratory Facilities (Secondary Barriers)

1. Laboratory doors must be self closing with key card access (or other secure access method). Access to the laboratory is restricted to entry by a series of two self-closing doors. A clothing change room (anteroom) may be included in the passageway between the two self-closing doors.
2. Laboratories must have a sink for hand washing. The sink must be hands-free or automatically operated. It should be located near the exit door. If the laboratory is segregated into different laboratories, a sink must also be available for hand washing in each zone.
3. The laboratory must be designed so that it can be easily cleaned and decontaminated. Seams, floors, walls, and ceiling surfaces should be sealed. Spaces around doors and ventilation openings should be sealed to facilitate space decontamination.
    a. Floors must be slip resistant, impervious to liquids, and resistant to chemicals.
    b. Walls should be constructed to produce a sealed smooth finish that can be easily cleaned and decontaminated.
    c. Ceilings should be constructed, sealed, and finished in the same general manner as walls.
    d. Decontamination of the entire laboratory will be considered when there has been gross contamination of the space, significant changes in laboratory
    e. usage, for major renovations, or maintenance shut downs. Selection of the appropriate materials and methods used to decontaminate the laboratory must be based on the situation and biological agents in use.
4. Laboratory furniture must be capable of supporting anticipated loads and uses.
5. Spaces between benches, cabinets, and equipment must be accessible for cleaning.
    a. Bench tops must be impervious to water and resistant to heat, organic solvents, acids, alkalis, and other chemicals.
    b. Chairs used in laboratory work must be covered with a non-porous material that can be easily cleaned and decontaminated with appropriate disinfectant.
6. All windows in the laboratory must be sealed.

7. BSCs must be installed so that fluctuations of the room air supply and exhaust do not interfere with proper operations. BSCs should be located away from doors, heavily traveled laboratory areas, and other possible airflow disruptions.
8. Vacuum lines must be protected with HEPA filters, or their equivalent. Filters must be replaced as needed. Filters must be properly disposed.
9. If floor drains are present, an appropriate liquid disinfectant in the trap will be required with associated maintenance schedule documentation.
10. An eyewash station must be readily available in the laboratory.
11. A ducted air ventilation system is required. This system must provide sustained directional airflow by drawing air into the laboratory from "clean" areas toward "potentially contaminated" areas. The laboratory shall be designed such that under failure conditions the airflow will not be reversed.
    a. Laboratory personnel must be able to verify directional air flow. A visual monitoring device which confirms directional air flow must be provided at the laboratory entry. Audible alarms should be considered to notify personnel of air flow disruption.
    b. The laboratory exhaust air must not re-circulate to any other area of the building.
    c. The laboratory building exhaust air should be dispersed away from occupied areas and from building air intake locations or the exhaust air must be HEPA filtered.
12. BSCs can also be connected to the laboratory exhaust system by either a thimble (canopy) connection or a direct (hard) connection. Provisions to assure proper safety cabinet performance and air system operation must be verified. BSCs will be certified at least annually by an outside contractor to assure correct performance
13. A method for decontaminating all laboratory wastes should be available in the facility, preferably within the laboratory (e.g., autoclave, chemical disinfection, incineration, or other validated decontamination method).
14. Equipment that may produce infectious aerosols must be contained in devices that exhaust air through HEPA filtration before being discharged into the laboratory. These HEPA filters should be tested and/or replaced at least annually.
15. Enhanced environmental and personal protection may be required by the agent summary statement, risk assessment, or applicable local, state, or federal regulations. These laboratory enhancements may include, for example, one or more of the following; an anteroom for clean storage of equipment and supplies with dress-in, shower-out capabilities; gas tight dampers to facilitate laboratory isolation; final HEPA filtration of the laboratory exhaust air; laboratory effluent decontamination; and advanced access control devices such as biometrics. HEPA filter housings should have gas-tight isolation dampers; decontamination ports; and/or bag-in/bag-out (with appropriate decontamination procedures) capability. The HEPA filter housing should allow for leak testing of each filter and assembly. The filters and the housing should be certified at least annually.
16. The BSL-3 facility design, operational parameters, and procedures must be verified and documented prior to operation. Facilities must be re-verified and documented at least annually.

IV. Transporting and Shipping

Outgoing shipments of hazardous materials, biological, chemical, and/or radioisotopes, must be coordinated through RMS. Shippers of hazardous materials must have documented DOT/IATA/ICAO mandated training. The University Mail Room will not accept hazardous materials shipments. Transporters will not pick up any hazardous materials without specific Hazardous Materials documents which are available through RMS. Packaging should be approved by RMS before use. Please call (334) 844-4805 for further information. Packaging and shipping requirements can be found in Appendix C of the BMBL.

NOTE: There are significant PERSONAL fines/possible imprisonment associated with improper packaging and shipping of hazardous materials.

Transport of Hazardous Materials by Research Personnel

Diagnostic and clinical specimens, infectious materials, and rDNA molecules need to be packaged in a sealed, leak proof, primary container (e.g., glass tube), which is securely positioned in a secondary leak proof and closable container (e.g., cooler, ice chest) clearly labeled with the biohazard symbol. A list of contents as well as emergency information (e.g., PI's phone number) needs to accompany the material (e.g., attached to the cooler in a plastic pouch).

The use of private cars for the transportation of such materials on or off campus is highly discouraged. University vehicles are available upon request through individual departments.

In case of an emergency (e.g., car accident), contact 911 and inform them of the presence of biohazardous materials and contact RMS if able.

Import/Export of Etiologic Agents

Import/export requirements for etiologic agents are included in Appendix C of the BMBL. Please contact RMS at 844-4870 for more information.

Transport of Select Agents

Any transport of Select Agents off campus requires a Commercial Drivers License (CDL). Please contact RMS for more information at 844-4870.

V. Select Agent Program

In United States law, select agents are pathogens or biological toxins which have been declared by the U.S. Department of Health and Human Services or by the U.S. Department of Agriculture to have the "potential to pose a severe threat to public health and safety". The Centers for Disease Control and Prevention administers Auburn University's Select Agent Program. The CDC regulates the laboratories which may possess, use, or transfer select agents within the United States. For more information regarding the Select Agent Program please visit the CDC website and contact RMS at 844-4870.

VI. Roles and Responsibilities

The consideration of biological safety issues is a campus-wide concern. With the ever-broadening interest in genetic engineering, agricultural production, and food safety, the need for standard biological safety policies and procedures is critical. A clear, consistent understanding of the organisms involved is an important first step in program organization.

This manual provides general guidelines and operating procedures for research involving the use of biological agents and toxins at Auburn University. All work involving these materials shall be performed in accordance with this manual. In addition the following materials shall be subject to the provisions of this manual.

Generation of rDNA - All experiments involving the generation and/or use of rDNA require IBC registration and may require approval by the IBC. The National Institutes of Health (NIH) Guidelines for Research Involving Recombinant DNA Molecules is the definitive reference for rDNA research in the United States. There may be rDNA experiments which are not covered by the guidelines that require review and approval by outside agencies before initiation or funding. These experiments are common in the agricultural and environmental sciences. If the experimental protocol is not covered by the guidelines, contact RMS at (334) 844-4870 for determination of further review.

If you have specific questions about a particular host-vector system not covered by the guidelines, please contact the Office of Recombinant DNA Activities (ORDA), National Institutes of Health at (301) 496-9838 or FAX to (301) 496-9839. Updates to the NIH rDNA guidelines are published in the Federal Register and are available on the internet at the NIH website.

Human Research - All research conducted by faculty staff or students that involves the use of human subjects must be reviewed and approved under Institutional Review Board (IRB) procedures prior to the involvement or recruitment of any subject.

Use of Animals - All animal experiments involving the use of rDNA; infectious or transmissible agents; human blood, body fluids, tissues, or toxins must be submitted to both the IBC and Institutional Animal Care and Use Committee (IACUC) for review and approval. IACUC guidelines are available from the Office of Animal Resources. Investigators who are uncertain how to categorize agents should contact RMS.

Transgenic Animals and Plants - Experiments to genetically engineer animals and plants require IBC registration. The NIH rDNA guidelines provide specific biocontainment recommendations for experiments involving the creation and/or use of genetically engineered animals and plants.

Other Research - Work involving chemicals or radioactive materials shall comply with the requirements of the Chemical Hygiene Plan, and/or the Radiation Safety Manual available on the AU Risk Management and Safety web page.

All work involving the use of recombinant DNA and genetically modified organisms shall be performed in accordance with the NIH Guidelines for Research Involving rDNA Materials.

The latest edition of the Biosafety in Biomedical and Microbiological Laboratories (BMBL) is an integral component of the Auburn University Biological Safety Manual. All descriptions of containment and handling of specific agents contained in the BMBL have been adopted as Auburn University.

Any work with human pathogens shall comply with the Auburn University Exposure Control Plan.

Institutional Biosafety Committee

The Institutional Biosafety Committee (IBC) is charged by the President of Auburn University to formulate policy and procedures related to the use of biohazardous agents, including: human, animal, and plant pathogens, other infectious agents, toxins, and recombinant DNA (rDNA). As mandated by the NIH, experiments involving human gene therapy, formation of transgenic animals or plants, and the generation and/or use of rDNA must be reviewed and approved by the IBC. Auburn University also requires IBC review and approval for use of RG2 or higher biohazardous agents.

IBC Organization Structure

The President of Auburn University will appoint members to the IBC and designate one member to serve as chairperson. In order to provide the quality of input needed for in-depth consideration of research activities presenting real or potential hazards, the membership shall be composed of faculty, laboratory staff, community members, and continuing members. The specific composition and membership terms of the IBC shall be as follows: For more information on the composition of the IBC please go to: IBC. All IBC members are required to sign an annual Confidentiality Agreement for service on the committee.

Responsibilities

The IBC has the responsibility of assessing risks and potential environmental impact associated with investigations involving biological agents and will make recommendations for safe conduct of such studies. The IBC functions on behalf of the institution to ensure that experimental work is performed in compliance with applicable policies, guidelines, and regulations for work with biohazardous materials and agents.

The IBC does not monitor activities which are appropriately the concern of other established institutional groups, e.g., Radiological Safety Committee, Institutional Review Board, and Institutional Animal Care and Use Committee. However, the IBC will closely interact with these groups in a concerted effort to minimize health risks for Auburn University personnel, students, and the general public.

All requests for action by the IBC shall be put in writing and submitted via the OVPR Office of Compliance. The registration document is the Biological Use Authorization Form (BUA). The BUA and instructions for completion may be found on the Risk Management and Safety web page.

Associate Director of Risk Management and Safety

The Associate Director of RMS is primarily responsible for implementation of the biological safety program. Day-to-day management of the program may be delegated to others within RMS, including the Biological Safety Officer. Major duties or activities of the Associate Director include the following:

- Serve as secretary of IBC
- Monitor compliance with university safety practices and procedures regarding potentially infectious and biohazardous materials.
- Assist in the preparation and periodic updating of a biosafety manual which is in accordance with University policy and consistent with government regulatory guidelines.

Biological Safety Officer

NIH guidelines require that a Biological Safety Officer be appointed whenever an institution engages in rDNA research at either BL-3 or large scale as defined by NIH. The Biological Safety Officer (BSO) is responsible for the day-to-day management of the biological safety program.

Responsibilities include:

- Assumption of duties and activities listed for the Associate Director of Risk Management and Safety. Serve as a continuing member of the IBC.
- Provide consultation to investigators on matters relating to laboratory safety, appropriate handling and containment of biohazardous agents, decontamination, and disposal of biohazardous wastes.
- Aid investigators in the development of appropriate emergency measures for dealing with accidental spills and contamination.
- Conduct surveillance of laboratories in which biohazardous agents are employed to ensure compliance with approved protocol, prescribed safety guidelines, safety training, and correction of deficiencies.
- Investigate incidents involving biohazardous agents to determine probable cause and identify any violation of the approved protocol, safety guidelines, or breech of containment. Upon completing the investigation, the Associate Director of RMS will prepare a report of findings for review and action by the IBC. Copies will be provided to the Vice President for Research. Reports involving rDNA will be forwarded to the NIH.
- Monitor intra-campus transport to ensure compliance with the rigorous containment procedures described herein. Provide information for off-campus shipment of biohazardous materials.

- Serve on building and renovation committees. Review plans for new facilities and modifications of existing structures where potentially pathogenic microorganisms; etiologic agents of RG2, RG3; infectious agents; chemical carcinogens; and/or rDNA materials will be used.
- Develop and conduct training programs for laboratory personnel using biohazardous agents, specifically to promote techniques for the safe handling and disposal of biohazardous materials.
- Serve as liaison between the university and outside regulatory agencies concerned with the use of biohazardous agents.
- Coordinate the off-site treatment of infectious wastes.
- Coordinate with the PI on the close out of a laboratory when a protocol using potentially pathogenic microorganisms is completed, when the PI moves laboratories or when the investigator leaves the university. These agents include:
    - etiologic agents of RG2, RG3
    - infectious agents
    - human tissue and blood borne pathogens
    - organisms containing rDNA.

Principal Investigators

Ultimate responsibility for the safe conduct of research involving biohazardous agents rests with the PI. The PI shall comply with the requirements contained in this manual and implement all necessary precautions to prevent undesirable consequences of experimental work conducted within the laboratory.

The PI shall:

- monitor daily operations of the laboratory
- inform/train persons who enter the laboratory
- file a BUA and obtain written approval of the IBC before performing work involving the use of biohazardous agents and toxins
- establish standard operation and decontamination procedures
- establish emergency procedures
- report any incident involving spill/release, injury, exposure, etc.
- arrange for immunization and/or health surveillance of laboratory personnel if deemed appropriate for the research project
- cooperate with the Biosafety Officer and other members of RMS during inspection visits, provide correction of noted deficiencies.

Department Heads/Chairpersons, Research Institute and Center Directors

The Department Heads/Chairpersons, Research Institute and Center Directors of each department, research institute, center or unit is responsible for the general safety of faculty, staff, and students in his/her overall area. The chief administrator shall insure that prior to initiation of work; each PI using a biohazardous agent files the appropriate registration forms.

The Department Heads/Chairpersons, Research Institute and Center Directors is mutually responsible, along with the PI, for informing the IBC of work involving biohazardous agents and for reporting accidents or incidents to the Biological Safety Officer.

The Department Heads/Chairpersons, Research Institute and Center Directors insures that students have received instruction regarding safety procedures in teaching laboratories or field situations where biohazardous agents are used.

The Department Heads/Chairpersons, Research Institute and Center Directors is responsible for providing appropriate facilities and safety equipment for proposed research or instruction involving biohazardous agents.

The Department Heads/Chairpersons, Research Institute and Center Directors is mutually responsible, along with the faculty members, for supervising teaching laboratories, informing students of proper precautions for working with pathogenic microorganisms, and for assuring that proper precautions are taken.

Laboratory Personnel

All laboratory personnel shall adhere to established safety practices/guidelines.

Laboratory personnel are responsible for keeping themselves informed of the risks involved with working in a laboratory and the risks associated with the specific agents they will be working with. Laboratory personnel will participate in laboratory safety training and are encouraged to request additional training if they feel they are unequipped to deal safely with the risks.

Laboratory personnel shall report all unsafe practices to the PI.

Laboratory personnel must report all accidents and injuries to the PI.

Administrative Controls

Review and Approval of Biohazardous Studies

The IBC can effectively carry out its designated functions only if it has adequate prior knowledge of potentially hazardous research projects. Therefore, all instructional, research, and outreach projects involving potentially pathogenic microorganisms; etiologic agents of RG2, RG3; infectious agents; oncogenic viruses; human tissue and bloodborne pathogens; and in vitro construction or propagation of rDNA molecules, must be reviewed and approved in writing by the IBC.

It is not the purpose of the IBC to pass judgment on scientific merits, or to consider "risk" versus "expected benefits" of potentially hazardous research. Rather, it is the concern of this committee that the safety precautions proposed

for the experimental work are adequate for the protection of personnel and environment. In general, the review process will focus on (1) qualifications of the PI, (2) agents to be employed, (3) risks presented by experimental procedures, (4) adequacy of containment equipment and facilities, (5) training level of persons directly associated with the work, (6) the need for health surveillance of laboratory personnel, and (7) other factors relevant to safe conduct of the study.

The PI and the IBC must concur on all matters relating to containment requirements, safe practices, and handling and disposal procedures for biohazardous agents. In the event of nonconcurrence, recommendations of the IBC shall prevail until such time as they are modified or rescinded by appellate decision of an administrative review. Administrative review may include outside reviewers, chaired by the Vice President for Research. Questions relating to rDNA studies not covered by the NIH Guidelines will be referred to the NIH Office of Recombinant DNA Activities for resolution.

RG3 Agents normally require BL3 containment due to the potential risk to workers and the environment. Therefore, the IBC will perform a more stringent evaluation procedure for oversight of this type of work. The IBC reserves the right to request full justification of all RG3 protocols including issues of scientific merit, compliance with regulatory requirements (including all surveillance, data reporting and adverse event reporting requirements; required containment levels for the research; assessment of the facilities, procedures, practices and training and expertise of personnel involved in the research; and risk/benefit of these projects.)

Agents or organisms or biohazardous materials requiring BL4 containment are not permitted in Auburn University facilities.

Clinical and Diagnostic Labs

Clinical and diagnostic labs are exempt from the BUA process.

Archival Samples/Cultures

Some laboratories maintain cultures and/or archival samples of biohazardous agents. In these instances, a generic BUA for maintenance and storage of reference, client and other samples/cultures should be submitted. An inventory of agents should be attached and updated whenever significant changes are made. Significant changes include additions or deletions of additional species or strains of inventoried agents that demonstrate a need for more stringent containment. If the responsible PI or individual utilizes any of those same agents in a specific teaching, research or outreach project, an additional BUA must be submitted.

Select Agent Use

PIs wishing to utilize CDC Select Agents must contact RMS prior to any work or acquirement of agents.

Biological Use Authorization (BUA)

All requests for action by the IBC shall be put in writing and submitted using the Biological Use Authorization Form (BUA). The BUA and instructions for completion are available on the Risk Management and Safety web page. Standard Operating Procedures for the BUA are included as Appendix A. Registrations approved by the IBC will be active for three years from the date of approval. Within the last quarter of the third year, the PI will be given the opportunity to renew the registration for an additional three years. After six years have elapsed, the investigator will be required to submit a new BUA for review and approval by the IBC.

The IBC shall meet as often as necessary to conduct business, typically on a monthly basis. It will be the responsibility of the chairperson to: (1) determine if additional meetings are needed; and (2) officially inform appropriate person(s) of the actions of the committee. The Secretary of the IBC has the responsibility to see that a complete file is kept of minutes, documents, and reports received.

It is important for faculty and staff members to understand that certain information, as described below, may be subject to public scrutiny under a disclosure provision of current NIH guidelines and applicable state and federal laws. This disclosure provision requires Auburn University, upon request, to make available to the public, all minutes of IBC meetings pertaining to recombinant DNA activities, and any documents or reports submitted or received from federal funding agencies, which the latter are required to make public e.g., Memoranda of Understanding and Agreement, reports of guidelines violations and significant research related accidents, facility inspection reports, and agency directives to modify projects (NIH Guidelines for Research Involving Recombinant DNA Materials)..

In accordance with NIH guidelines, Auburn University will forward public comments regarding IBC actions and the IBC response to those public comments to the NIH.

Stop Work Procedures

The Associate Director of RMS, with concurrence from the Chair of the IBC, or with concurrence of three (3) members of the IBC if the Chair is unavailable, may stop any work with microbial agents or any hazardous research project that creates an unreasonable hazard to personnel or involves experiments prohibited by the University. The entire Committee then will review the situation and will complete the review within a working week and

forward written recommendation(s) to the President of Auburn University or his designee, the Vice President for Research, for final action which will occur within two weeks of receipt of the recommendations. A copy of the final action determination will be maintained in the researcher's BUA file.

Planned/Unplanned Shutdowns

The BSO must be notified immediately in the event of:

- any mechanical malfunction
- any systems breakdown
- shutdowns of any nature
- preventative maintenance of primary containment equipment or components.

In the case of an unplanned event and if Facilities Division staff is not already on the scene, the BSO or the Associate Director of RMS will notify appropriate Facilities Division staff. Proper precautions must be taken immediately. All experiments must be halted and the biological agents secured (e.g., containers sealed or containers placed in freezer or refrigerator). The area must be cordoned off during the entire time of the shut down. No further activities will be allowed until the BSO certifies that the facility is safe to use.

Biohazard Warning Signs and Posting

Each laboratory must clearly display signage that provides safety information to visitors and service personnel. Signs must contain designations for all laboratory hazards in use within the laboratory (carcinogens, acutely toxic agents, reproductive hazards, biohazards, radioactive materials, lasers, and magnetic fields). The RMS Door Sign Program will be utilized for all laboratory doors. Contact RMS for more information.

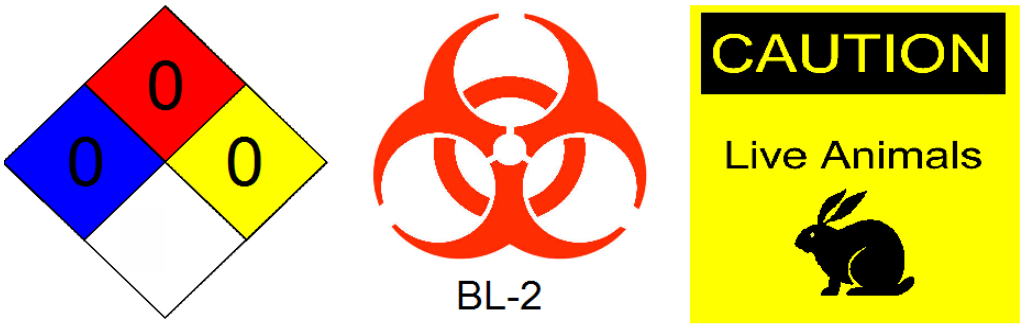
Appropriate Personal Protective Equipment required for entry into the lab shall be specified on the biohazard sign.

Figure 1 – Door Sign Example

7/31/2009

NOTICE

AUTHORIZED PERSONNEL ONLY



CONTACTS FOR Leach Science Center 201	NAME	BUILDING	OFFICE	PHONE #	HOME #
For entry or information:	John Smith	Leach Science Center	201A	4-1234	235-1235
In emergency:	John Smith Jr	Leach Science Center	201	4-1254	235-1234
In emergency:	John Smith Sr.	Leach Science Center	201	4-1254	235-1236

All areas and laboratories which contain biohazardous or toxic agents must clearly display signs stating "EATING, DRINKING, SMOKING AND APPLYING COSMETICS ARE PROHIBITED IN THIS AREA."

Occupational Safety and Health Program

Personnel having substantial animal contact shall participate in the Occupational Safety and Health/Medical Monitoring Program, administered through the Office of Animal Resources.

Vaccines will be offered to all clearly identified “at-risk personnel” where the benefits clearly exceed the risk. Immunoprophylaxis may provide an additional level of protection. For more information see Appendix B of the BMBL. Vaccination costs will be the responsibility of the home department.

For a more detailed explanation of this program, refer to the University’s Office of Animal Resources Occupational Health and Safety Program.

Bloodborne Pathogens

All human cell cultures, human blood, and body fluids are considered to be potentially contaminated and are treated as RG2 agents and must be handled at BL-2 or above.

The AU Exposure Control Plan provides guidelines and procedures for personnel who are occupationally at-risk of exposure to bloodborne pathogens. In accordance with this plan, a Hepatitis B vaccination will be made available, at no cost to the individual (charged to home department). Post-exposure evaluation and follow-up will also be provided.

For a more detailed explanation of this program, refer to the University's Exposure Control Plan.

Waste Management

Infectious and Biological Waste Management

Disposal of medical waste, infectious waste, autoclave bags, pipettes, sharps, and biological waste must be performed in accordance with Alabama Department of Environmental Management (ADEM) medical waste regulations. Auburn University has developed, "A Guide to the Handling and Disposal of Medical Waste," that lists compliance procedures and University policies.

Only specified autoclave bags and sharps disposal containers may be used.

NOTE: All cultures of microorganisms, including RG1 agents, shall be inactivated, using appropriate procedures, before disposal. This is considered good laboratory practice.

Appendix A – Standard Operating Procedures

SOP 1 – Biohazard Spill Clean Up Procedures
SOP 2 – General Laboratory Definitions and Procedures
SOP 3 – Biological Safety Cabinets (BSCs)
SOP 4 – Personal Protective Equipment (PPE)
SOP 5 – Decontamination Procedures

SOP 1 – Biohazard Spill Clean Up Procedures

The following procedures are provided as a guideline to biohazardous spill cleanup. Additional information regarding emergency plans is in Appendix F of the BMBL.

Inside The Biological Safety Cabinet (BSC):

Wear lab coat, safety goggles, and gloves during cleanup.

- Allow cabinet to run during cleanup.
- Apply disinfectant and allow a minimum of 20 minutes contact time.
- Wipe up spillage with disposable disinfectant-soaked cloth.
- Wipe the walls, work surface, and any equipment in the cabinet with a disinfectant-soaked cloth.
- Discard contaminated disposable materials in an appropriate biohazardous waste container(s) and autoclave before discarding IAW A Guide to the Handling and Disposal of Medical Waste.
- Place contaminated reusable items in autoclave bags, autoclavable pans with lids, or wrap in newspaper before autoclaving and before cleanup.
- Expose non-autoclavable materials to disinfectant, 20 minute contact time, before removal from the BSC.
- Remove protective clothing used during cleanup and place in an autoclave bag for autoclaving.
- Run cabinet 10 minutes after cleanup before resuming work or turning cabinet off.

In The Lab, Outside The BSC

Clear the area of all personnel. Wait for aerosol to settle before entering spill area. The time required will depend on ventilation within the area, but a general rule of thumb is 30 minutes. Remove any contaminated clothing and place in autoclave bag to be autoclaved. Wear a disposable gown, safety goggles, and gloves at all times when pathogenic organisms are present. Initiate cleanup with disinfectant as follows:

- Soak paper towels in disinfectant and place over spill.
- Encircle the spill with additional disinfectant, being careful to minimize aerosolization while ensuring adequate contact.
- Decontaminate all items within spill area.
- Allow 20 minutes contact time to ensure germicidal action of disinfectant.
- Wipe equipment with 1:10 part bleach solution, followed by water, and then a 70% alcohol solution.
- Place disposable contaminated spill materials in a biohazardous waste container(s) appropriate for autoclaving.
- Place contaminated reusable items in autoclave bags, autoclavable pans with lids, or wrap in newspaper before autoclaving and cleanup.

Inside the centrifuge

If a centrifuge tube breaks while the centrifuge is running, turn off the motor and allow the machine to rest for 30 minutes before opening. If breakage is discovered after the machine has stopped, close the lid immediately and allow the unit to rest for 30 minutes.

- Unplug centrifuge before initiating clean-up.
- Don strong, thick, rubber gloves and other Personal Protective Equipment (PPE) before proceeding with clean-up.
- Flood centrifuge bowl with germicidal disinfectant. Place paper towels soaked in disinfectant over the entire spill area. Allow 20 minutes of contact time.
- Use mechanical means, such as forceps, to remove broken tubes and glass fragments. Place broken tubes and glass in sharps container for autoclaving and disposal as infectious waste IAW A Guide to the Handling and Disposal of Medical Waste.
- Remove buckets, trunnions, and rotor then place in disinfectant for 24 hours or autoclave. Place disinfectant soaked paper towels over equipment if transport is necessary.
- Unbroken, capped tubes may be placed in disinfectant and recovered after 20 minute contact time, or these tubes can be autoclaved.
- Use mechanical means to remove remaining disinfectant soaked materials from centrifuge bowl and discard as infectious waste IAW Appendix D.
- Place disinfectant soaked paper towels in the centrifuge bowl and allow soaking overnight. Wipe down again with disinfectant, wash with water, and dry. Discard disinfectant soaked materials as infectious waste IAW Appendix D.
- Remove protective clothing used during cleanup and place in a biohazard bag for autoclaving. Wash hands after removing gloves.

Outside the Lab, During Transport

Transport biohazardous material in an unbreakable, well-sealed, primary container placed inside an unbreakable, lidded container labeled with the biohazard symbol (cooler, plastic pan or pail).

Should a spill occur in a public area, do not attempt to clean it up without appropriate PPE.

As an interim measure, with gloved hands, place disinfectant soaked paper towels directly on spilled materials to prevent spread of contamination. To assure adequate contact, surround the spill with disinfectant, if available, taking care to minimize aerosols.

Call RMS at (334) 844-4870 for assistance with cleanup. Call 911 for assistance if spill takes place outside of normal working hours.

SOP 2 – General Laboratory Definitions and Procedures

Personal Protective Equipment (PPE)

PPE such as proper gloves, safety glasses, and a laboratory coat must be worn whenever biological work is conducted in the laboratory. No sandals or open-toed shoes are allowed in the laboratory. Long pants/skirt are recommended.

Handwashing

Hands must be washed immediately or as soon as feasible after removing gloves or other personal protective clothing.

Use of Sharps

Minimize the use and exposure to sharps in the workplace. Never recap, bend, or shear needles. When possible, replace glassware with less damaging materials such as plastic. Keep sharps containers readily available in all locations where sharps waste may be generated.

Clean Areas

Eating, drinking, smoking, applying cosmetics or lip balm, and handling contact lenses in the laboratory are prohibited in work areas where there is a reasonable likelihood of occupational exposure.

Aerosol Generation

Any procedures that could potentially generate aerosols or other inhalation hazards must be performed in a manner that will minimize airborne pathogen transmission.

Proper Labeling

Place a color-coded label incorporating the universal biohazard label on the work surface of any potentially contaminated equipment or work surface to warn others of biohazard contamination which may not be easily visible. This includes freezers, refrigerators, and incubators.

Autoclave Safety

Always wear heat-resistant gloves, goggles or safety glasses, and a laboratory coat when opening an autoclave. Be sure to allow the superheated steam to exit before attempting to remove the contents.

Spills

Always clean spills from the periphery of the spill towards the center. All cleaning materials must be disposed of in an appropriate manner. See SOP 1.

Mouth Pipetting

Mouth pipetting may lead to accidental ingestion of biological specimens and is strictly prohibited.

Decontamination Procedures

A 0.5% sodium hypochlorite (a freshly prepared 1:10 dilution of household bleach) must be used to decontaminate equipment and work surfaces. In locations where bleach would cause corrosion, an iodophor (e.g., Wescodyne) must be used to decontaminate.

Local Transport of Infectious Materials

All infectious materials transported to and from the laboratory must be enclosed in a primary container with sealed lid, which must then be enclosed in a secondary leak-proof, non-breakable container, appropriately labeled with the biohazard symbol. Any specimens transported to and from off-campus satellite facilities must be escorted by a responsible lab employee. All transport must be performed in a university vehicle.

Storage

All infectious materials to be stored must be clearly labeled with the universal biohazard symbol. The storage space (e.g., freezer, refrigerator) must also be similarly labeled.

Bloodborne Pathogens

All PI's using human or non-human primate blood or blood products, unfixed tissue, body fluids, or organ or cell cultures of human or non-human primate origin, must follow the procedures outlined in the Auburn University Bloodborne Pathogen Exposure Control Plan.

Human Organ and Cell Culture

All PI's using human organ or cell cultures (primary cultures, cell strains, cell lines), must handle all such cultures under BL2 conditions and in accordance with the Bloodborne Pathogen Standard, unless the IBC has specifically approved a lower standard of containment. IRB approval may be needed for use of these materials.

Transport of “Select Agents”/Toxins

RMS must be notified of all transfers.

Waste Disposal

All biological waste must be autoclaved before disposal in accordance with Auburn University's Medical Waste Management Guide. Blue autoclave bags are to be used for this purpose.

Injuries

All injuries and accidental autoinoculation, ingestion or inhalations of infectious agents, must be reported immediately to the lab director or supervisor and RMS (344) 844-4870. Affected employees should be sent to the Auburn University Medical Clinic (AUMC) for evaluation, possible treatment and/or referral. Outside AUMC hours of operation, send affected employees to East Alabama Medical Center Emergency Room. Dial 911 immediately for any medical emergency.

Shipments

All domestic and international shipments of biological materials must follow University policy and all applicable federal and international regulations. Proper permits/licenses must be obtained before importing or exporting biological materials. Contact RMS prior to assist with shipments.

Emergency Preparedness

In case of natural disasters, fires, or power failure the following precautions must be taken:

- In power failures, immediately discontinue all work until power is restored. If a tissue culture hood is being used, then all open containers must be closed, gas turned off, and hood sash closed.
- In natural disasters, personnel must immediately follow standard emergency procedures (911; 4-4870 (RMS)). Upon return to facility, Personal Protective Equipment (PPE) must be used when entering a lab to decontaminate any disaster-related release of infectious material. Contain released material using spill procedures. Emergency personnel should don PPE before entering lab and/or areas housing infected animals.
- In case of fire, personnel must immediately follow standard emergency procedures (911; 4-4870 (RMS)). Temperatures sufficient to ignite materials will inactivate infectious agents used in the laboratory. However, emergency personnel should don PPE before entering the lab and follow disinfecting procedures described above for decontaminating any released infectious materials not involved in the fire.

Pipettes and Pipetting Aids

Pipettes are used for volumetric measurements and transfer of fluids that may contain infectious, toxic, corrosive, or radioactive agents. Laboratory associated infections have occurred from oral aspiration of infectious materials, mouth transfer via a contaminated finger, and inhalation of aerosols. Exposures to aerosols may occur when liquid from a pipette is dropped onto the work surface, when cultures are mixed by pipetting, or when the last drop of an inoculum is blown out. A pipette may become a hazardous piece of equipment if improperly used. The safe pipetting techniques which follow are required to minimize the potential for exposure to hazardous materials.

- Never mouth pipette. Always use a pipetting aid.
- If working with biohazardous or toxic fluid, confine pipetting operations to a biosafety cabinet.
- Always use cotton plugged pipettes when pipetting biohazardous or toxic materials, even when safety pipetting aids are used.
- Do not prepare biohazardous materials by bubbling expiratory air through a liquid with a pipette.
- Do not forcibly expel biohazardous material out of a pipette.
- Never mix biohazardous or toxic material by suction and expulsion through a pipette.

- When pipetting, avoid accidental release of infectious droplets. Place a disinfectant soaked towel on the work surface and autoclave the towel after use.
- Use "to deliver" pipettes rather than those requiring "blowout."
- Do not discharge material from a pipette at a height. Whenever possible allow the discharge to run down the container wall.
- Place contaminated, reusable pipettes horizontally in a pan containing enough liquid disinfectant to completely cover them. Do not place pipettes vertically into a cylinder.

Discard contaminated disposable pipettes in an appropriate sharps container. Autoclave the container when it is 2/3 to 3/4 full and dispose as medical waste. See "A Guide to the Handling and Disposal of Medical Waste," Appendix D. Call (344) 844-4870 if you need an additional copy. This document is also available on the RMS web page. Pans or sharps containers for contaminated pipettes should be placed inside the biosafety cabinet, if possible.

Syringes and Needles

Syringes and hypodermic needles are dangerous instruments. The use of needles and syringes should be restricted to procedures for which there is no alternative. Blunt cannulas should be used as alternatives to needles wherever possible (i.e., procedures such as oral or intranasal animal inoculations). Needles and syringes should never be used as a substitute for pipettes. When needles and syringes must be used, the following procedures are recommended:

- Use disposable needle locking syringe units whenever possible.
- When using syringes and needles with biohazardous or potentially infectious agents:
- Work in a biosafety cabinet whenever possible.
- Wear gloves.
- Fill the syringe carefully to minimize air bubbles.
- Expel air, liquid, and bubbles from the syringe vertically into a cotton pledget moistened with disinfectant.
- Do not use a syringe to mix infectious fluid forcefully.
- Do not contaminate the needle hub when filling the syringe. This will help avoid transfer of infectious material to fingers.
- Wrap the needle and stopper in a cotton pledget moistened with disinfectant when removing a needle from a rubber-stoppered bottle.
- Bending, recapping, clipping, or removing needles from syringes is prohibited. The use of needle nipping devices is prohibited.
- Use a separate pan of disinfectant for reusable syringes and needles. Do not place them in pans containing pipettes or other glassware to eliminate sorting later.
- Used disposable needles and syringes must be placed in appropriate sharps disposal containers and discarded as medical waste. See "A Guide to the Handling and Disposal of Medical Waste", or Appendix D.

Cryostats

Frozen sections of unfixed tissue infected with an etiologic agent pose a risk because accidents can occur. Freezing tissue does not necessarily inactivate infectious agents. Freezing propellants under pressure should not be used for frozen sections as they may cause spattering of droplets of infectious materials. Gloves should be worn during preparation of frozen sections. When working with biohazardous material in a cryostat, the following is recommended:

- Consider the contents of the cryostat to be contaminated and decontaminate it frequently with 70% ethanol.
- Consider trimmings and sections of tissue that accumulate in the cryostat to be potentially infectious and remove them during decontamination.
- Defrost and decontaminate the cryostat with a tuberculocidal hospital disinfectant once a week and immediately after tissue known to contain bloodborne pathogens, Mycobacterium tuberculosis or other infectious agents is cut.
- Handle microtome knives with extreme care, stainless steel mesh gloves should be worn when changing knife blades.
- Consider solutions for staining potentially infected frozen sections to be contaminated.

Centrifuge Equipment

Hazards associated with centrifuging include mechanical failure and the creation of aerosols. To minimize the risk of mechanical failure, centrifuges must be maintained and used according to the manufacturer's instructions. Users should be properly trained and operating instructions that include safety precautions should be prominently posted on the unit.

Aerosols are created by practices such as filling centrifuge tubes, removing plugs or caps from tubes after centrifugation, removing supernatant, and resuspending sedimented pellets. The greatest aerosol hazard is created if a tube breaks during centrifugation. To minimize the generation of aerosols when centrifuging biohazardous material, the following procedures should be followed:

- Use sealed tubes and safety buckets that seal with O-rings. Before use, inspect tubes, O-rings, and buckets for cracks, chips, erosions, bits of broken glass, etc. Do not use aluminum foil to cap centrifuge tubes because it may detach or rupture during centrifugation.
- Fill and open centrifuge tubes, rotors, and accessories in a BSC. Avoid overfilling of centrifuge tubes so that closures do not become wet. After tubes are filled and sealed, wipe them down with disinfectant.
- Add disinfectant to the space between the tube and the bucket to disinfect material in the event of breakage during centrifugation.
- Always balance buckets, tubes, and rotors properly before centrifugation.
- Do not decant or pour off supernatant. Use a vacuum system with appropriate in-line reservoirs and filters. For more information, call RMS at (344) 844-4870.
- Work in a BSC when resuspending sedimented material. Use a swirling rotary motion rather than shaking. If shaking is necessary, wait a few minutes to permit the aerosol to settle before opening the tube.

- Small low-speed centrifuges may be placed in a BSC during use to reduce aerosol escape. High-speed centrifuges pose additional hazards. Precautions should be taken to filter the exhaust air from vacuum lines, to avoid metal fatigue resulting in disintegration of rotors and to use proper cleaning techniques and centrifuge components. Manufacturers' recommendations must be meticulously followed to avoid metal fatigue, distortion and corrosion.
- Avoid the use of celluloid (cellulose nitrate) tubes with biohazardous materials. Celluloid centrifuge tubes are highly flammable and prone to shrinkage with age. They distort on boiling and can be highly explosive in an autoclave. If celluloid tubes must be used, an appropriate chemical disinfectant must be used to disinfect them.

Blenders, Ultrasonic Disrupters, Grinders and Lyophilizers

The use of blenders, ultrasonic disrupters, grinders, and/or lyophilizers devices may result in considerable aerosol production. Blenders, grinders, and cell-disruption equipment should be used in a BSC when working with biohazardous materials.

Safety blenders, although expensive, are designed to prevent leakage from the bottom of the blender jar, provide a cooling jacket to avoid biological inactivation and to withstand sterilization by autoclaving. If blender rotors are not leakproof, they should be tested with sterile saline or dye solution prior to use with biohazardous material. The use of glass blender jars is not recommended; however, if they must be used, glass jars should be covered with a polypropylene jar to prevent spraying of glass and/or contents if the jar breaks. A towel moistened with disinfectant should be placed over the top of the blender during use. Before opening the blender jar, allow the unit to rest for at least one minute to allow the aerosol to settle. The device should be decontaminated promptly after use.

Lyophilizers and ampoules

Depending on lyophilizer design, aerosol production may occur when material is loaded or removed from the unit. If possible, sample material should be loaded in a BSC. The vacuum pump exhaust should be filtered to remove any hazardous agents or, alternatively, the pump can be vented into a BSC. After lyophilization is complete, all surfaces of the unit that has been exposed to the agent should be disinfected. If the lyophilizer is equipped with a removable chamber, it should be closed off and moved to a BSC for unloading and decontamination. Handling of cultures should be minimized and vapor traps should be used wherever possible.

Opening ampoules containing liquid or lyophilized culture material should be performed in a BSC to control the aerosol produced. Gloves must be worn. To open, nick the neck of the ampoule with a file, wrap it in disinfectant soaked towel, hold the ampoule upright and snap it open at the nick. Reconstitute the contents of the ampoule by slowly adding liquid to avoid aerosolization of the dried material. Mix the contents without bubbling and withdraw it into a fresh container. Discard the towel and ampoule top and bottom as infectious waste.

Ampoules used to store biohazardous material in liquid nitrogen have exploded causing eye injuries. The use of polypropylene tubes eliminates this hazard. These tubes are available dust-free or presterilized and are fitted with polyethylene caps and silicone washers. Heat sealable polypropylene tubes are also available.

Loop Sterilizers and Bunsen Burners

Sterilization of inoculation loops or needles in an open flame generates small-particle aerosols which may contain viable microorganisms. The use of a shielded electric incinerator minimizes aerosol production during loop sterilization. Alternatively, disposable plastic loops and needles may be used for culture work where electric incinerators or gas flames are not available. The loops are semiquantitative and can be used for counting bacteria.

Continuous flame gas burners should not be used in BSCs. These burners can produce turbulence which disturbs the protective airflow patterns of the cabinet. Additionally, the heat produced by the continuous flame may damage the HEPA filter. If a gas burner must be used, one with a pilot light should be selected.

Housekeeping

Good housekeeping in laboratories is essential to reduce risks and protect the integrity of biological experiments. Routine housekeeping must be relied upon to provide work areas free of significant sources of contamination. Housekeeping procedures should be based on the highest degree of risk to which personnel and experimental integrity may be subjected.

Laboratory personnel are responsible for cleaning laboratory benches, equipment, and areas that require specialized technical knowledge. Additional laboratory housekeeping concerns include:

- Keeping the laboratory neat and free of clutter; surfaces should be clean and free of infrequently used chemicals, glassware, and equipment. Access to sinks, eyewashes, emergency showers, and fire extinguishers must not be blocked.
- Proper disposal of chemicals and waste - old and unused chemicals should be disposed of promptly and properly. Call RMS at (344) 844-4805/4870 for details.
- Providing a workplace that is free of physical hazards - aisles and corridors should be free of tripping hazards. Attention should be paid to electrical safety, specifically; as it relates to the use of extension cords, proper grounding, avoidance of overloaded electrical circuits, and the creation of electrical hazards in wet areas.
- Removing unnecessary items from floors, under benches, or in corners.
- Properly securing all compressed gas cylinders.
- Never using fume hoods for storage of chemicals or other materials.
- Practical custodial concerns include:
- Dry sweeping and dusting which may lead to the formation of aerosols is prohibited.
- The usual wet or dry industrial type vacuum cleaner is a potent aerosol generator and, unless equipped with High Efficiency Particulate Air (HEPA) filter, must not be used in the biological research laboratory. Use of these industrial type vacuums

that do not have HEPA filters, is prohibited in order to protect personnel as well as the integrity of the experiment. Wet and dry units with HEPA filters on the exhaust are available from a number of manufacturers.

SOP 3 – Biological Safety Cabinets (BSCs)

Biological Safety Cabinets (BSCs) - BSCs are designed to contain aerosols generated during work with infectious material through the use of laminar air flow and high efficiency particulate air (HEPA) filtration. Consult CDC/NIH BMBL for a discussion of the types and uses of BSCs.

Safe and Effective Use of Biosafety Cabinets

In general:

- Ensure your BSC is certified when it is installed, after it is moved, and annually thereafter. For information on cabinet certification, call RMS at (344) 844-4870. Check the magnahelic gauge regularly for indication of a problem.
- Understand how your cabinet works.
- Do not disrupt the protective airflow pattern of the BSC. Such things as rapidly moving your arms in and out of the cabinet, people walking rapidly behind you, and/or open lab doors may disrupt the airflow pattern and reduce the effectiveness of the BSC.
- Plan your work.

- Minimize the storage of materials in and around the BSC.
- Always leave the BSC running.

Operational directions:

- Before using, wipe work surface with 70% alcohol. Wipe off each item you need for your procedures and place in cabinet.
- DO NOT place objects over the front air intake grille. DO NOT block the rear exhaust grille.
- Segregate contaminated and clean items. Work from "clean to dirty."
- Place a pan with disinfectant and/or a sharps container inside the BSC for pipette discard. DO NOT use vertical pipette discard canisters on the floor outside cabinet.
- It is not necessary to flame items; this creates turbulence in airflow and will compromise sterility; heat buildup may damage the filters.
- Move arms slowly when removing or introducing new items into the BSC.
- If you use a piece of equipment that creates air turbulence in the BSC (such as a centrifuge, blender) place equipment in the back 1/3 of the cabinet; stop other work while equipment is operating.
- Protect the building vacuum system from biohazards by placing a HEPA cartridge filter or its equivalent between the vacuum trap and the source valve in the cabinet.
- Clean up all spills in the cabinet immediately. Wait 10 minutes before resuming work.
- When work is finished, remove all materials and wipe all interior surfaces with 70% alcohol.
- Remove lab coat and wash hands thoroughly before leaving laboratory.

SOP 4 – Personal Protective Equipment (PPE)

Safety equipment includes items for personal protection such as gloves, coats, gowns, shoe covers, boots, respirators, face shields, safety glasses or goggles. PPE is often used in combination with BSCs and other devices which contain the biohazardous agents, animals or materials. When it is impractical to work in BSCs, PPE may form the primary barrier between personnel and infectious materials. Examples include certain animal studies, animal necropsy, agent production activities, and activities relating to maintenance, service or support of the laboratory facility.

PPE is used to protect personnel from contact with hazardous materials and infectious agents. Appropriate clothing may also protect the experiment from contamination. PPE must be provided without cost to personnel. The following PPE is recommended for regular use:

Face Protection

Goggles in combination with masks, or chin length face shields, or other splatter guards are required whenever there is the possibility of splashes, sprays, or splatters of infectious or other hazardous materials to the face.

Laboratory clothing

This category includes: laboratory coats, smocks, scrub suits, and gowns. Long sleeved garments should be used to minimize the contamination of skin or street clothes, and to reduce shedding of microorganisms from the arms. In circumstances where it is anticipated that splashes may occur, the garment must be resistant to liquid penetration (in order to protect clothing from contamination). If the garment is not disposable, it must be capable of withstanding sterilization in the event it becomes contaminated. Additional criteria for selecting clothing are: comfort, appearance, closure types, and location, antistatic properties, and durability. Protective clothing must be removed and left in the laboratory before leaving for non-laboratory areas. Disposables should be available for visitors, maintenance, and service workers in the event it is required. All protective clothing should be either discarded in the laboratory or laundered by the facility. Personnel must not launder laboratory clothing at home.

Gloves

Gloves must be selected based on the hazards involved and the activity to be conducted. Gloves must be worn when working with biohazards, toxic substances and other physically hazardous agents. Temperature resistant gloves must be worn when handling hot material or dry ice. Delicate work requiring a high degree of precision dictates the use of thin walled gloves. Protection from contact with toxic or corrosive chemicals may also be required. For assistance in glove selection consult the selection information in Prudent Practices for Handling Hazardous Chemicals in Laboratories. For further assistance call RMS at (334) 844-4870.

When working with hazardous materials, the lower sleeve and the cuff of the laboratory garment should be overlapped by the glove. A long sleeved glove or disposable arm-shield may be worn for further protection of the garment.

In some instances “double gloving” may be appropriate. If a spill occurs, hands will be protected after the contaminated outer gloves are removed. Gloves must be disposed of when contaminated and removed when work with infectious material is completed. Gloves are not to be worn outside the laboratory. Disposable gloves must not be washed or reused.

Respirators

In certain instances additional PPE may be required. Respirator selection is based on the hazard and the protection factor required. Personnel who require respiratory protection must contact RMS for inclusion in the Auburn University Respiratory Protection Program. The program provides: a physical examination to ensure no health conditions exist that would be exacerbated by respirator usage; annual fit testing to ensure proper respirator size and type; and training to ensure proper respirator use and maintenance. Under no circumstances shall anyone wear a respirator unless he/she is a participant in the program.

Contact RMS for assistance in selection of respirators or other personal protective equipment.

Laundry

All personal protective clothing must be cleaned, laundered and disposed of by the employer at no cost to employees. Apparel contaminated with blood or other potentially infectious

materials should be handled as little as possible and decontaminated, preferably by autoclaving, before being sent to the laundry for cleaning. Appropriate PPE must be worn by employees who handle contaminated laundry.

SOP 5 – Decontamination Procedures

Decontamination is a term used to describe a process or treatment that renders a medical device, instrument, or environmental surface safe to handle. A decontamination procedure can range from sterilization to simple cleaning with soap and water. Sterilization, disinfection, and antisepsis are all forms of decontamination.

Sterilization is the use of a physical or chemical procedure to destroy all microbial life, including highly resistant bacterial endospores.

Disinfection eliminates virtually all pathogenic non-sporeforming microorganisms, but not necessarily all microbial forms on inanimate objects (work surfaces, equipment, etc.). Effectiveness is influenced by the kinds and numbers of organisms, the amount of organic matter, the object to be disinfected, and the chemical exposure time, temperature, and concentration.

Antisepsis is the application of a liquid antimicrobial chemical to skin or living tissue to inhibit or destroy microorganisms. It includes swabbing an injection site on a person or animal and hand washing with germicidal solutions. Although some chemicals may be utilized as either a disinfectant or an antiseptic, adequacy for one application does not guarantee adequacy for the other. Manufacturers' recommendations for appropriate use of germicides should always be followed.

General Procedures: All infectious materials and all contaminated equipment or apparatus should be decontaminated before being washed, stored, or discarded. Autoclaving is the

preferred method. Each individual working with biohazardous material should be responsible for its proper handling.

Biohazardous materials should not be placed in autoclaves overnight in anticipation of autoclaving the next day.

Autoclaves should not be operated unattended or by untrained personnel.

Special precautions should be taken to prevent accidental removal of material from an autoclave before it has been sterilized or simultaneous opening of both doors on a double door autoclave.

Dry hypochlorites, or any other strong oxidizing material, must not be autoclaved with organic materials such as paper, cloth or oil.

OXIDIZER + ORGANIC MATERIAL + HEAT = POSSIBLE EXPLOSION

Methods: There are four main categories of physical and chemical decontamination. They are heat, liquid disinfection, vapors and gases, and radiation. Each category is discussed briefly below.

Heat

Wet heat is the most dependable method of sterilization. Autoclaving (saturated steam under pressure of approximately 15 PSI to achieve a chamber temperature of at least 250º F for a prescribed time) is the most convenient method of rapidly achieving destruction of all forms of microbial life. In addition to proper temperature and time, prevention of entrapment of air is critical to achieving sterility. Material to be sterilized must come in contact with steam and heat. Chemical indicators, e.g. autoclave tape, must be used with each load placed in the autoclave. The use of autoclave tape alone is not an adequate monitor of efficacy. Autoclave sterility monitoring should be conducted on a regular basis using appropriate biological indicators (B. stearothermophilus spore strips) placed at locations throughout the autoclave. The spores, which can survive 250º F for 5 minutes but are killed at 250º F in 13 minutes, are more resistant to heat than most, thereby providing an adequate safety margin when validating decontamination procedures. Each type of container employed should be individually tested with these spores because efficacy varies with the load, fluid volume, etc. Autoclaves used for sterilization of materials subject to ADEM Medical Waste Rules must comply with those rules. See "A Guide to the Handling and Disposal of Medical Waste", Appendix D for those requirements.

Dry Heat is less efficient than wet heat and requires longer times and/or higher temperatures to achieve sterilization. It is suitable for the destruction of viable organisms on impermeable non-organic surfaces such as glass, but it is not reliable in the presence of shallow layers of organic or inorganic materials which may act as insulation. Sterilization of glassware by dry heat can usually be accomplished at 160 - 170º C for periods of 2 - 4 hours. Dry heat sterilizers should be monitored on a regular basis using appropriate biological indicators [Bacillus subtilis (globigii) spore strips].

Incineration is another effective means of decontamination by heat. As a disposal method incineration has the advantage of reducing the volume of the material prior to its final disposal.

Liquid disinfection - The most practical use of liquid disinfectants is for surface decontamination, and when used in sufficient concentration, to decontaminate liquid wastes prior to final disposal in the sanitary sewer. If liquid disinfectants are used, they must have been shown to be effective against the organism(s) present.

Liquid disinfectants are available under a wide variety of trade names. In general, these can be classified as halogens, acids, alkalis, heavy metal salts, quaternary ammonium compounds, phenolic compounds, aldehydes, ketones, alcohols, and amines. The more active a compound is, the more likely it is to have undesirable characteristics such as corrosivity. No liquid disinfectant is equally useful or effective under all conditions and for all viable agents. Properties of common disinfectants can be viewed at, http://www.ehrs.upenn.edu/programs/bio/bsm/Table3.xls.

Vapors and gases

A variety of vapors and gases possess decontamination properties. Vapors and gases are primarily used to decontaminate biological safety cabinets and associated systems, bulky or stationary equipment not suited to liquid disinfectants, instruments or optics which might be damaged by other decontamination methods, rooms, buildings, and associated air-handling systems. Agents included in this category are: glutaraldehyde and formaldehyde vapor, ethylene oxide gas, peracetic acid, and hydrogen peroxide vapor. When used in closed systems and under controlled conditions of temperature and humidity, excellent disinfection can be obtained. Great care must be taken during use because of the hazardous nature of many of these compounds. Contact RMS for monitoring requirements if these compounds are to be used.

Radiation

Although ionizing radiation will destroy microorganisms, it is not a practical tool for laboratory use. Nonionizing radiation in the form of ultraviolet radiation (UV) is used for inactivating viruses, bacteria, and fungi. It will destroy airborne microorganisms and inactivate microorganisms on exposed surfaces or in the presence of products of unstable composition that cannot be treated by conventional means.

Because of the low penetrating power of UV, microorganisms inside dust or soil particles will be protected from its action, limiting its usefulness. UV is used in air locks, animal holding areas, ventilated cabinets and laboratory rooms to reduce levels of airborne microorganisms and maintain good air hygiene. Because UV can cause burns to the eye and skin of people exposed for even a short period of time, proper shielding should be maintained when it is in use. UV lamps that are used for space decontamination should be interlocked with the general room or cabinet illumination, so that turning on the lights extinguishes the UV.

UV lamps are not recommended for decontamination unless they are properly maintained. Because UV lamp intensity or destructive power decreases with time, it should be checked with a UV meter yearly. Frequent lamp cleaning (at least every few weeks) is necessary to prevent accumulation of dust and dirt which drastically reduces its effectiveness. If UV must be used, it should be used when areas are not occupied.

Appendix B - Recommended Biosafety Levels for Infectious Agents

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

Appendix C - Plant Biosafety Level Criteria

Research involving rDNA-containing plants, plant-associated microorganisms, and small animals shall be conducted in accordance with Appendix P. Physical And Biological Containment For Recombinant DNA Research Involving Plants as contained in the latest copy of the document Guidelines For Research Involving Recombinant DNA Molecules (NIH Guidelines) published by the National Institutes of Health (NIH).

The NIH Guidelines specify physical and biological containment conditions and practices suitable to the greenhouse conduct of experiments involving rDNA-containing plants, plant-associated microorganisms, and small animals. These guidelines supersede standard rDNA containment requirements when research plants are of a size, number, or have growth requirements that preclude the use of standard laboratory containment conditions. Plants covered in the NIH guidelines include, but are not limited to, mosses, liverworts, macroscopic algae, and vascular plants including terrestrial crops, forest, and ornamental species. Plant-associated microorganisms include viroids, virusoids, viruses, bacteria, fungi, protozoans, certain small algae, and microorganisms that have a benign or beneficial association with plants, such as; certain Rhizobium species and microorganisms known to cause plant diseases. These guidelines apply to microorganisms which are modified with the objective of fostering an association with plants. Plant-associated small animals include those arthropods that:

- are in obligate association with plants,
- are plant pests,
- are plant pollinators, or
- transmit plant disease agents.

Other small animals include nematodes for which tests of biological properties necessitate the use of plants. Microorganisms associated with such small animals (e.g., pathogens or symbionts) are included.

Researchers handling similar organisms (plants, plant-associated microorganisms, and small animals) for which a USDA, APHIS, U.S. Department of the Interior or U.S. Public Health Service permit is required must notify the IBC of its research by memo with a copy of the permit attached.

Appendix D – Infectious Waste Management

All cultures of microorganisms should be inactivated before disposal. This includes cultures of RG1 organisms. This is considered good laboratory practice.

Infectious Waste Management

Consult A Guide to the Handling and Management of Medical Waste for complete details on the disposal of infectious waste in accordance with ADEM regulations and University policy. A link to the most recent copy is included below. Additional copies are available by request, either call (344) 844-4870 or send a request to RMS, 316 Nuclear Science Center.

Mixed Waste

Mixed wastes are potentially infectious waste contaminated with other types of waste, e.g., radioisotopes or toxic/carcinogenic compounds. Because of the difficulty in disposal of wastes regulated by more than one set of requirements and more than one regulatory agency, it is critical that provision be made for proper management prior to the initiation of any research that might result in mixed waste. Mixed wastes may require special containers, labeling, storage, etc. Contact RMS prior to initiation of any research that might result in potentially infectious waste with multiple hazards.

Animals

The Institutional Animal Care and Use Committee must approve the disposal method for research animals and animal parts that are considered to be infectious waste. RMS will be consulted when necessary.

Appendix E Training

As described in Section VI, the PI is responsible for the training of everyone working in his/her laboratory. Training is possibly the single most important action a PI can take to promote a safe and healthy working environment/ laboratory. All training must be documented.

At a minimum, training will consist of the following:

- Basic Laboratory Safety Training
- Laboratory - Specific Biosafety Training
- Medical Waste Management Training
- Bloodborne Pathogens Training (if necessary)

Violations of established safety procedures resulting in accident/incident may be followed by recommendations from the IBC that would require additional specialized training. These recommendations would be routed through the Vice President for Research, as well as appropriate deans, directors and department heads.

Basic Laboratory Safety Training consists of a series of short videotapes that can be viewed in approximately one hour. The videotape cassettes can be borrowed from RMS. Alternatively, the tapes can be viewed at that office. Please call (344) 844-4870 to request the tapes or schedule a viewing. The required tapes are:

Practicing Safe Science	29 minutes
Chemical Hazards	10 minutes
Emergency Response	12 minutes
Centrifugation Hazards	12 minutes

Specific Laboratory Biosafety Training will be provided by the PI and will include generalized training for the biosafety level at which the laboratory operates and specialized training for specific hazards present in that laboratory. This training shall be reviewed annually.

Medical Waste Management Training is provided by the RMS, without cost. Please call 4-4870 to schedule Medical Waste Management Training. This training must be documented.

Bloodborne Pathogens Training is necessary for any student or employee who works with human blood, human blood components, and products made from human blood, human organs, or human body fluids. Bloodborne Pathogens Training is provided by RMS. Please call (344) 844-4870 to schedule Bloodborne Pathogens Training. This training must be documented.

Violations of established safety procedures resulting in accident/incident may be followed by recommendations from the IBC that would require additional specialized training. These recommendations would be routed through the Vice President for Research, as well as appropriate deans, directors and department heads.

DOCUMENTATION

All training must be documented, to ensure each laboratory worker receives the required training, and to provide written testimony to that effect. Auburn University has developed an example form that may be used for this purpose. Besides documentation, the form provides the PI with a checklist of safety training that might be required by hazards present within his/her laboratory.

The Training Form, signature pages from the memoranda described above and any additional training certifications should be kept together in a central location within each laboratory. The PI is responsible for ensuring that the required training is provided and documented as discussed in Section II of this manual. Documentation will be examined during regular laboratory inspections performed by the RMS.

For the purposes of this program a certified training course is a course that is offered through RMS or a recognized, independent source of training.

Teaching Laboratories

Instructors in teaching laboratories should provide specific training for the hazards expected to be encountered in the laboratory procedures utilized. Because of the brevity of courses, the large number of students involved, and the detail and length of the Training Form, it is recommended that each instructor develop a streamlined training form emphasizing the particular elements required for their course. Both the instructor and student should sign the training form.