THE UNIVERSITY OF NORTH FLORIDA

BIOSAFETY MANUAL

Revised June 2008

FOREWORD

It is the policy of the University of North Florida to provide a safe working environment. The primary responsibility for ensuring safe conduct and conditions in the laboratory rests with the Lab Instructor, Lab Supervisor or Lab Manager.

This manual, developed by the Department of Environmental Health, Safety, Insurance and Risk Management (EH&S), is intended to offer guidance in the area of biological teaching and research. It is a part of the overall effort to provide a comprehensive safety and health program for the University community.

This manual is available to laboratory personnel upon request and in the offices of EH&S and the Natural Sciences Laboratory Manager. Impacted laboratory personnel must know and follow the procedures outlined in the manual. All operations performed in the laboratory must be planned and executed in accordance with this document. In addition, each employee is expected to develop safe personal habits to reduce the likelihood of injuries and exposures.

This document was developed in compliance with state and federal regulations including, the Florida Department of Health, the Department of Environmental Protection (DEP), the Occupational Safety and Health Administration (OSHA) and the Environmental Protection Agency (EPA). The departments of Physical Facilities, Facilities Planning, and EH&S will assist lab personnel in keeping facilities and procedures employed in the laboratory compatible with this manual. This manual will be reviewed at least annually and is readily available to employees and their representatives.

EMERGENCY CONTACTS

UNIVERSITY POLICE DEPARTMENT

Information: 620-2804
Emergency: 620-2800 or 911 from campus phone

CHEMICAL ACCIDENT

Environmental Health & Safety: 620-2019
University Police Department: 620-2800 or 911 from campus phone

BIOLOGICAL ACCIDENT

Environmental Health & Safety: 620-2019
University Police Department: 620-2800 or 911 from campus phone

FOR ADDITIONAL INFORMATION CONTACT:

Division of Safety
National Institutes of Health
9000 Rockville Pike
Building 31, Room 1C-02
Bethesda, MD 20892
(301) 496-1357

Office of Biosafety
Centers for Disease Control
Atlanta, GA 30333
(404) 639-3883

National Animal Disease Control
US Department of Agriculture
Ames, Iowa 50010
(515) 239-8200

Biotechnology Permits (General Information)
Animal & Plant Health Inspection Service
US Department of Agriculture
Room 850, Federal Building
6505 Belcrest Road
Hyattsville, Md 20782
(301) 436-7601

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I. Introduction

The University of North Florida Biological Safety Manual provides guidance to instructors and researchers working with biological agents. It should be used in conjunction with the UNF Chemical Hygiene Plan, Biowaste Plan, and Bloodborne Pathogens Plan, where appropriate. The recommendations of this manual are applicable to teaching, training, clinical, diagnostic, research, and other laboratory activities in which viable microorganisms or clinical materials are used.

II. PRINCIPLES OF BIOSAFETY

The term "containment" is used in describing methods for managing infectious agents in the laboratory environment where they are being handled or maintained. The purpose of containment is to reduce exposure of laboratory employees, other persons and the outside environment to potentially hazardous or infectious agents. The three elements of containment include laboratory practices and techniques, safety equipment and facility design. Primary containment, the protection of personnel and the immediate laboratory environment from exposure to infectious agents, is provided by good micro-biological technique and the use of appropriate safety equipment. Secondary containment, the protection of the external laboratory environment from exposure to infectious materials, is provided by a combination of facility design and operational practices.

A. Laboratory Practice and Technique

The most important element of containment is strict adherence to standard microbiological practices and techniques. Persons working with infectious agents or infected materials must be aware of potential hazards and be trained and proficient in the practices and techniques required for handling such material safely. The lab manager and lab supervisor is responsible for providing or arranging for appropriate training of personnel with assistance from EH&S.

Additional measures may be necessary when standard laboratory practices are not sufficient to control the hazard associated with a particular agent or laboratory procedure. The selection of additional safety practices is the responsibility of the lab manager and lab supervisor and must be commensurate with the inherent risk associated with the agent or procedures. An example of an additional practice would be a vaccination requirement. (See Appendix 5 for vaccination recommendations)

B. Safety Equipment

Such equipment includes biological safety cabinets and a variety of enclosed containers. The biological safety cabinet (BSC) is the principal device used to provide containment of infectious aerosols generated by many microbiological procedures.

Three types of BSC's (Class I, II, III) used in microbiological labs are described in Appendix 6. Open-fronted Class I and Class II BSC's are partial containment cabinets which offer significant levels of protection to laboratory personnel and to the environment when used in conjunction with good microbiological techniques. The gas-tight Class III BSC provides the highest attainable level of protection to personnel and the environment. An example of an enclosed container is the capped centrifuge bottle which prevents the release of aerosols during centrifugation.

Safety equipment also includes items for personal protection such as gloves, coats, gowns, shoe covers, boots, respirators, face shields, and safety glasses. These personal protective devices are often used in

combination with biological safety cabinets and other devices which contain the agents, animals and materials being worked with. In some situations in which it is impractical to work in biological safety cabinets, personal protective devices may form the primary barrier between personnel and the infectious materials. Examples include certain animal studies, animal necropsy, production activities and activities relating to maintenance, service or support to the laboratory.

C. Facility Design

The design of the facility is critical for providing protection to persons outside the laboratory and in the community in the event that an infectious agent is accidentally released in the laboratory. It is the responsibility of the lab manager, EH&S, Physical Facilities and Facilities Planning to provide lab facilities commensurate with the function of the laboratory. The following information describes three facility designs, in ascending order of containment level.

1. Basic Laboratory

This laboratory provides general space appropriate for work with defined viable agents which are not associated with disease processes in healthy adults or which do not colonize in humans. All activities are regularly conducted on the open bench using standard laboratory practices.

2. Containment Laboratory

This laboratory provides general space appropriate for work with infectious agents or potentially infectious materials when the hazard levels are low and laboratory personnel can be adequately protected by standard laboratory practices. Work is commonly conducted on the open bench with certain operations confined to BSC's. Conventional laboratory designs are adequate. Areas known to be sources of general contamination such as animal rooms and waste staging areas should not be adjacent to media, processing areas, tissue culture laboratories or patient care activities. Public areas and general offices to which non-laboratory staff require frequent access should be separated from spaces which primarily support laboratory functions.

3. High Containment Laboratory

This laboratory has special engineering features which make it possible for laboratory workers to handle hazardous materials without endangering themselves, the community or the environment. The unique features which distinguish this laboratory from the basic and containment laboratories are the provisions for access control, a specialized ventilation system and vacuum line isolation (See Appendix 6).

The high containment laboratory may be an entire building or a single module or complex of modules within a building. In all cases, the laboratory is separated by a controlled access zone from areas open to the public and laboratory personnel.

III. BIOSAFETY LEVELS

A. Introduction

This section describes the various biosafety levels which consist of combinations of laboratory practices and techniques, safety equipment and laboratory facilities which are commensurate with the potential hazard posed by the infectious agents used in the laboratory.

Section B describes three general biosafety levels which are appropriate for most experimentation.

Section C describes three working levels, to be followed when using experimental animals. They are designated Animal Biosafety Levels. The levels to which the various agents are assigned can be found in Section D.

Please note that work with agents above biosafety level 3 is handled on a case by case basis. Please contact EH&S for regulations regarding this type of experimentation.

The lab supervisors and instructors are directly responsible for the safe operation of the laboratory. Their knowledge and judgment are critical in assessing risks and appropriately applying the recommendations of these guidelines. The recommended biosafety level to which a particular agent is assigned represents minimal conditions under which the agent can ordinarily be safely handled. Special characteristics of the agents used, training and experience of personnel and the nature or function of the laboratory may further influence the supervisor in applying these guidelines.

Work with known agents should be conducted at the biosafety level recommended unless specific information is available to suggest that virulence, pathogenicity, antibiotic resistance patterns, etc., are significantly altered to require more or allow less stringent practices to be used. As a general policy, clinical, field and environmental specimens should be handled at the level recommended for most pathogenic agents the clinical diagnosis or other evidence suggests is likely to be present. For example, sputa submitted for tuberculosis examination should be handled from the outset as potentially infectious (biosafety level 3). Personnel working with specimens or tissues submitted for rabies examination should be immunized and take appropriate precautions to prevent parenteral or aerosol exposures.

Personnel working with specimens or tissues of domestic and wild animals should be aware of known or potential zoonotic infections and should be immunized if vaccines are available (see the Agent Listing and Appendix 5 for vaccination recommendations). Personnel wishing to work at a biosafety level lower than that specified in this manual are required to have prior approval to do so. The request, with supporting materials should be submitted to EH&S.

B. General Specifications

Biosafety Level I (BL1) is suitable for experiments involving agents of no known or of minimal potential hazard to laboratory personnel and the environment. The laboratory is not separated from the general traffic patterns of the building. Work is generally conducted on open bench tops and special containment equipment is not required or generally used. Laboratory personnel have specific training in the procedures conducted in the laboratory and are supervised by personnel with general training in microbiology or a related science. BL 1 practices, safety equipment and facilities are those appropriate for undergraduate and secondary educational training and teaching laboratories and for other facilities working with defined and characterized strains of viable microorganisms not known to cause disease in healthy adult humans or not known to colonize in humans. (Bacillus cereus, Naegleria fowleri, and canine distemper virus are representative of those microorganisms assigned to BL 1.)

Note however, that many agents not ordinarily associated with disease processes or colonization in humans are opportunistic pathogens and may cause infection for the young, the aged and for immuno-suppressed or immuno-incompetent individuals. Vaccine strains which have undergone multiple in vivo passages should not a priori be considered avirulent.

Biosafety Level I

1. Standard Microbiological Practices

a. Laboratory doors are kept closed when experiments are in progress.

b. Work surfaces are decontaminated daily and after each spill of viable material. (See Section IV for a list of common disinfectants.)

c. All contaminated liquids or solid wastes are decontaminated before being disposed of or otherwise handled (See Section VI).

d. Mechanical pipetting devices are used; mouth pipetting is prohibited. Eating, drinking, smoking, storing of food and applying cosmetics are not permitted in the work area.

e. Persons will wash their hands after handling viable materials or animals and before they leave the laboratory.

f. All procedures must be carefully performed to minimize the creation of aerosols.

g. The wearing of laboratory coats, gowns or uniforms is strongly recommended.

2. Special Practices

a. Contaminated materials that are to be decontaminated at a site away from the laboratory shall be placed in a durable container which is sealed before leaving the lab.

b. An insect and rodent control program is in effect.

3. Containment Equipment

Special containment equipment is generally not required for manipulations of agents assigned to BL 1.

4. Laboratory Facilities

a. The laboratory should be designed so that it is easily cleaned.

b. Bench tops should be impervious to water and resistant to acids, alkalis, organic solvents, and moderate heat.

c. Each laboratory should contain a hand washing sink.

d. If the laboratory has windows that open, they should be fitted with screens.

e. An autoclave for decontamination of infectious laboratory wastes should be available in the same building with the laboratory. (See Appendix 9)

Biosafety Level 2 (BL2) is similar to BL 1 and is suitable for experiments involving agents of moderate potential hazard to personnel and the environment. It differs in that laboratory personnel have specific training in handling pathogenic agents, access to the laboratory is limited when experiments are being conducted and that procedures involving large volumes or high concentrations are conducted in BSC's or other physical containment equipment. BL 2 practices, equipment and facilities are those which are applicable to clinical, diagnostic, teaching and other facilities working with the broad spectrum of indigenous moderate-risk agents present in the community and associated with human disease of varying severity. [The Hepatitis agents (Hepatitis A, Hepatitis B, Hepatitis non A-non B), the Salmonellae, and Toxoplasma spp, are representative of micro-organisms assigned to this containment level.] Primary hazards to personnel working with these agents relate to accidental auto-inoculation or ingestion of infectious materials.

Procedures with high aerosol potential may predictably and significantly increase the risk of exposure and must be conducted in primary containment equipment or devices.

Biosafety Level 2

1. Standard Microbiological Practices

a. Laboratory doors are kept closed when experiments are in progress.

b. Work surfaces are decontaminated at least daily and after each spill of viable material. (See Section IV for a list of common disinfectants)

c. All contaminated liquids or solid waste are decontaminated before being disposed of or otherwise handled. (See Section VI)

d. Mechanical Pipetting devices are used; mouth pipetting is prohibited.

e. Eating, drinking, smoking, storing food and applying cosmetics are not permitted in the work area.

f. Persons must wash their hands after handling infectious materials or animals and before they leave the lab.

g. All procedures are conducted carefully to minimize the creation of aerosols.

h. Laboratory coats, gowns or uniforms must be worn in the laboratory, but must NOT be worn in non-laboratory areas when soiled or contaminated.

2. Special Practices

a. Contaminated materials that are to be decontaminated at a site away from the laboratory shall be placed in durable, leak proof containers which are sealed before being removed from the laboratory.

b. An insect and rodent control program is in affect.

c. The laboratory supervisor will assure that only persons who have been advised of the potential hazard and met any specific entry requirements (e.g. immunization) may enter the laboratory or animal rooms.

d. When infectious materials or infected animals are present in the laboratory or animal rooms, a hazard warning sign, incorporating the universal biohazard symbol, is posted on all laboratory and animal room access doors and on other items (i.e. equipment, containers, materials) as appropriate to indicate the presence of viable infectious agents. The hazard warning sign should identify the agent, list the name of the laboratory supervisor or other responsible person(s) and indicate any special requirements for entering the area (immunization, respirators, etc.).

e. Access to the Laboratory is limited by the laboratory supervisor when experiments are being conducted. In general, persons who are at increased risk of acquiring infection or for whom infection may be unusually hazardous are not allowed in the laboratory or animal rooms. Persons at increased risk may include children, pregnant women, and individuals who are immunodeficient or immunosuppressed. The lab manager or lab supervisor has the final responsibility for assessing each individual circumstance and determining who may enter or work in the laboratory.

f. Animals not involved in the experiment being performed are not permitted in the lab.

g. The use of hypodermic needles and syringes is restricted to gavage, parental injection, and aspiration

of fluids from laboratory animals and diaphragm vaccine bottles. Hypodermic needles and syringes are not used as a substitute for automatic pipetting devices in the manipulation of infectious fluids. Serial dilutions of infectious agents should not be done in diaphragm bottles with needles and syringes because of the hazards of autoinoculation and of aerosol exposure. Cannulas should be used instead of sharp needles whenever possible.

h. If activities of lesser biohazard potential are conducted in the laboratory concurrently with activities requiring BL 2, all activities will be conducted at BL 2.

i. Gloves should be worn for all procedures requiring the handling of infectious materials or infected animals.

j. All spills, accidents, and overt or potential exposures to infectious materials must be immediately reported to the laboratory supervisor. A written record must be prepared and maintained. Appropriate medical evaluation, surveillance, and treatment must be provided. (See Sections VII and VIII)

k. Safety or operational instructions which identify known and potential hazards and which specify practices and procedures to minimize or eliminate such risks should be prepared or adopted as necessary. Personnel must be advised of special hazards and are required to read and follow standard practices and procedures.

3. Containment Equipment

Biological safety cabinets (Class I, II, or III) (See Appendix 6) or other appropriate personal protective or physical containment devices are used whenever:

a. Procedures with a high potential for creating aerosols are conducted. These may include centrifugation, grinding, blending, vigorous shaking or mixing, sonic disruption, opening containers of infectious materials whose internal pressures may be different from ambient, intranasal inoculation of animals, and harvesting infected tissue from animals or eggs.

b. High concentrations or large volumes of infectious agents are used. Such materials may be centrifuged in the open laboratory if sealed heads or centrifuge safety cups are used and if they are opened only in a biological safety cabinet.

4. Laboratory Facilities

a. The laboratory should be designed so that it is easily cleaned.

b. Bench tops should be impervious to water and resistant to acids, alkalis, organic solvents, and moderate heat. The use of plastic-backed absorbent toweling over work surface is recommended.

c. Each laboratory should contain a hand washing sink.

d. If the laboratory has windows that open, they should be fitted with screens.

e. An autoclave for decontamination of infectious laboratory wastes should be available in the same building with the laboratory. (See Appendix 9)

Biosafety Level 3 (BL3) is suitable for experiments involving agents of high potential risk to personnel and the environment. Laboratory personnel have specific training in handling pathogenic and potentially lethal agents and are supervised by competent scientists who are experienced in working with these agents.

Access to the laboratory is controlled by the lab supervisor. The laboratory has special engineering and design features and physical containment equipment and devices. All procedures involving the manipulation of infectious material are conducted within biological safety cabinets or other containment devices or by personnel wearing appropriate personal protective clothing. Level three practices, safety equipment and facilities are those which are applicable to clinical, diagnostic, teaching, research or production facilities working with indigenous or exotic agents which may cause serious and potentially lethal infections.

(Mycobacterium Tuberculosis, St. Louis Encephalitis Virus, and Coxiella burnetti are representative of microorganisms assigned to this level.) Primary hazards to personnel working with these agents relate to auto-inoculation, ingestion, and exposure to infectious aerosols.

Biosafety Level 3

1. Microbiological Practices

a. Laboratory doors are kept closed when experiments are in progress.

b. Work surfaces are decontaminated at least daily and after each spill of viable material. (See Section IV)

c. All contaminated liquids or solid wastes are decontaminated before being disposed of or otherwise handled. (See Section VI)

d. Mechanical pipetting devices are used; mouth pipetting is prohibited.

e. Eating, drinking, smoking, storing food, and applying cosmetics are not permitted in the work area.

f. Persons must wash their hands after handling viable materials or animals and before they leave the laboratory.

g. All procedures are conducted carefully to minimize the creation of aerosols.

h. Laboratory clothing that protects street clothing (i.e. solid front or wrap-around gowns, scrub suits, coveralls, etc.) is worn in the laboratory. FRONT- BUTTON LABORATORY COATS ARE UNSUITABLE. Laboratory clothing is not to be worn outside of the laboratory and is decontaminated before being laundered.

2. Special Practices

a. All contaminated materials shall be decontaminated within the generating laboratory.

b. An insect and rodent control program is in effect.

c. The laboratory supervisor will assure that only persons who have been advised of the potential biohazard, meet any specific entry requirements (e.g. immunization) and comply with all entry and exit procedures may enter the laboratory or animal room.

d. When infectious materials or infected animals are present in the laboratory or animal rooms, a hazard warning sign incorporating the universal biohazard symbol, is posted on all laboratory and animal room access doors and on other items (i.e. equipment, containers, materials) as appropriate to indicate the presence of viable infectious agents.

The hazard warning sign should identify the agent, list the name of the laboratory supervisor or other responsible person(s) and indicate any special conditions of entry into the area (immunizations, respirators, etc.).

e. Access to the laboratory is controlled by the laboratory supervisor and is restricted to persons whose presence is required for program or support needs. Persons who are at increased risk if acquiring infection or for whom infection may be unusually hazardous are not allowed in the laboratory or animal rooms. Persons at increased risk may include children, pregnant women, and individuals who are immunodeficient or immunosuppressed. The lab supervisor has the final responsibility for assessing each individual circumstance and determining who may enter or work in the laboratory.

f. Animals and plants not related to the experiment being conducted are not permitted in the laboratory.

g. The use of hypodermic needles is restricted to gavage, parenteral injection, and aspiration of fluids from lab animals and diaphragm vaccine bottles. Hypodermic syringes are not used as a substitute for automatic pipetting devices in the manipulation of infectious fluids. Serial dilutions of infectious agents should not be done in diaphragm bottles with syringes due to the potential for autoinoculation and aerosol exposure.

h. If activities of lesser biohazard potential are conducted in the laboratory concurrently with activities requiring BL 3, all work will be conducted at BL 3.

i. Gloves are worn when handling infectious materials or animals. Gloves should be removed aseptically and autoclaved with other laboratory wastes before being disposed of.

j. All spills, accidents and overt or potential exposures to infectious materials must be immediately reported to the laboratory supervisor. A written report must be prepared and maintained. Appropriate medical evaluation, surveillance and treatment must be provided.

k. Safety or operational instructions which identify known and potential hazards and which specify practices and procedures to minimize or eliminate such risks should be prepared or adopted. Personnel should be advised of special hazards and must read and follow required practices and procedures.

l. Molded surgical masks or respirators are worn in rooms containing infected animals.

m. All activities involving infectious materials are conducted in biological safety cabinets or other physical containment devices. No work in open vessels is conducted on the open bench.

n. The work surfaces of biological safety cabinets and other containment equipment are decontaminated when an experiment is finished. The use of plastic-backed paper toweling on non-perforated work surfaces within biological safety cabinets facilitates clean up following the completion of activities.

o. Baseline serum samples should be collected and stored for all laboratory and other at-risk personnel. Additional serum specimens must be collected in cases of known or suspected exposure.

3. Biosafety Equipment

Biological safety cabinets or other physical containment equipment for devices are used for all procedures and manipulations involving infectious material.

4. Laboratory Facilities

a. The surfaces of walls, floors and ceilings are water resistant and can be easily cleaned. Openings in these surfaces are sealed or capable of being sealed to facilitate decontaminating the area.

b. Bench tops should be impervious to water and resistant to acids, alkalis, organic solvents and moderate heat.

c. A foot or elbow-operated hand washing sink is provided near each laboratory exit door.

d. Windows in the laboratory are closed and sealed.

e. An autoclave for decontamination of laboratory wastes is available within the laboratory. Infectious wastes which must be removed to another area in the same building for decontamination must be held and transported in a covered, leakproof container. (See Appendix 9)

f. The laboratory is separated from areas which are open to unrestricted traffic flow within the building. Separation is provided by either a double-door change room and shower or an airlock or other access facility which requires passage through two sets of doors to enter the laboratory. Access to the laboratory area is designed to prevent entrance of free-living arthropods.

g. Access doors to the laboratory are self closing and locking.

h. An exhaust air ventilation system is provided. This system creates directional airflow that draws air into the laboratory through the entry area.

The building exhaust system can be used for this purpose if the exhaust air is not recirculated to any other area of the building. Personnel must verify that proper directional airflow (into the laboratory) is achieved. However, air within the laboratory can be recirculated. The exhaust air from the laboratory is discharged directly to the outside or through the building exhaust system so that it is dispersed away from occupied buildings and air intakes. The exhaust air from the laboratory that does not come from the biological safety cabinet can be discharged to the outside without being treated.

i. In laboratories which have supply air systems, the supply air and exhaust air system are interlocked to assure inward airflow at all times

j. The HEPA-filtered exhaust air from Class I or Class II biological safety cabinets should be discharged directly to the outside or through the building exhaust system. Air may be recirculated within the laboratory only after it has been filtered through tested and certified cabinet exhaust HEPA filters. Exhaust air from Class III biological safety cabinets is to be discharged to the outside through a building exhaust air system. It is recommended that these cabinets be connected to this system to avoid any interference with the air balance of the cabinet or building exhaust system.

k. Biological safety cabinets must be certified annually.

E. Animal Specifications

These guidelines presuppose that laboratory animal facilities, operational practices, and quality of animal care meet applicable standards and regulations and that appropriate species selection has been made for animal experiments (e.g. Guide for the Care and Use of Laboratory Animals, HEW Publication No. (NIH) 78-23, Rev. 1978, and Laboratory Animal Welfare Regulations, 9 CFR, Subchapter A, Parts 1, 2 and 3).

Ideally, facilities for laboratory animals used for studies of infectious or noninfectious disease should be physically separated from other activities such as animal production and quarantine, clinical laboratories

and especially from facilities that provide patient care. Animal facilities should be designed and constructed to facilitate cleaning and housekeeping. A "clean corridor/dirty corridor" layout is very useful in reducing cross contamination. Floor drains should be installed in animal facilities only on the basis of clearly defined needs. If floor drains are installed, the drain trap should always contain water. This section describes three combinations of practices, safety equipment and facilities for experiments on animals infected with agents which produce or may produce human infection. These combinations provide increasing levels of protection to personnel and to the environment and are recommended as minimal standards for activities involving infected laboratory mammals.

Animal Biosafety Level 1

1. Standard Practices

a. Door to animal rooms are self-closing and are kept closed when experiments are in progress.

b. Work surfaces are decontaminated following use or spills of viable materials. (See Section IV)

c. Eating, drinking, smoking, and storing food are not permitted in animal rooms.

d. Personnel shall wash their hands after handling viable cultures and animals and before leaving the animal room.

e. All procedures are carefully conducted to minimize the creation of aerosols.

f. An insect and rodent control program is in effect.

2. Special Practices

a. Bedding materials from cages used for animals infected with agents transmissible to humans are decontaminated--preferably by autoclaving--before being discarded. Cages used for animals infected with agents transmissible to humans are washed and/or rinsed with water, heated to at least 180°F, for at least 20 minutes.

b. The wearing of laboratory coats, gowns or uniforms in the animal room is recommended. It is further recommended that laboratory coats worn in the animal room not be worn in other areas.

3. Biosafety Equipment

Special containment equipment is generally not required for animals infected with agents assigned to BL 1.

4. Animal Facilities

a. The animal facility should be designed and constructed to facilitate cleaning and housekeeping.

b. A hand washing sink is available in the animal facility.

c. If the animal facility has windows that open, they shall be fitted with screens.

d. The animal facility shall be provided with inward directional airflow and that exhaust air be discharged to the outside without being recirculated to other rooms.

Animal Biosafety Level 2

1. Standard Practices

a. Doors to animal rooms are self-closing and are kept closed when experiments are in progress.

b. Work surfaces are decontaminated following use or spills of viable materials. (See Section IV)

c. Eating, drinking, smoking, and storing of food are not permitted in animal rooms.

d. Personnel shall wash their hands after handling viable cultures and animals and before leaving the animal room.

e. All procedures are carefully conducted to minimize the creation of aerosols.

f. An insect and rodent control program is in effect.

2. Special Practices

a. Cages shall be autoclaved before bedding is removed and before they are cleaned and washed.

b. Laboratory coats, gowns or uniforms shall be worn in the animal room but must not be worn elsewhere.

c. Surgical-type masks shall be worn by all personnel entering animal rooms housing nonhuman primates.

d. Access to the animal room is restricted by the laboratory or animal facility supervisor to personnel who have been advised of the potential hazard and who need to enter on an approved basis when experiments are in progress. In general, persons who may be at increased risk of acquiring infection or for whom infection might be unusually hazardous are not allowed in the animal room. Persons at increased risk may include children, pregnant women, and individuals who are immunodeficient or immunosuppressed. The lab supervisor has the final responsibility for assessing individual circumstances and determining who may enter or work in the animal room.

e. The laboratory supervisor will assure that only persons who have been advised of the potential hazard and meet any specific requirements (e.g. immunization) may enter the animal room.

f. Hazard warning signs, incorporating the universal biohazard warning symbol are posted on access doors to animal rooms when materials containing or animals infected with agents assigned to BL 2 or higher are present. The hazard warning sign should identify agent(s) in use, list the name of the laboratory supervisor or other responsible person(s) and indicate any special conditions for entry into the animal room (e.g., immunization, respirators).

g. Gloves are worn by personnel handling animals when the hazard of contact infection exists. Forceps should be used when handling and inoculating small laboratory animals to further reduce exposures of personnel to bites, scratches or unnecessary contact with infected animals.

h. All waste from the animal rooms are appropriately decontaminated-- preferably by autoclaving-- before being disposed of. Infected animal carcasses are autoclaved before being disposed of in sealed, leakproof containers. (See Section VI)

i. The use of hypodermic needles and syringes is restricted to gavage, parenteral injection or aspiration of fluids from laboratory animals and diaphragm bottles.

Serial dilutions of infectious agents should not be done in diaphragm bottles with needles and syringes

because of the hazards of autoinoculation and of aerosol exposure. Canulas should be used instead of sharp needles whenever possible.

j. If floor drains are provided, the drain trap is always filled with water.

3. Containment Equipment

Biological safety cabinets, other physical containment devices and/or personal protective devices (e.g. respirators, face shields) are used whenever procedures with a high potential for creating aerosols are conducted. These may include necropsy of infected animals, harvesting of infected tissues or fluids from animals or eggs, intranasal inoculation of animals and manipulations of high concentrations or large volumes of infectious materials. (See Appendix 6)

4. Animal Facilities

a. The animal facility shall be designed and constructed to facilitate cleaning and housekeeping.

b. A hand washing sink shall be available in the room where infected animals are housed.

c. If the animal facility has windows that open, they shall be fitted with screens.

d. The animal facility shall be provided with inward directional airflow and that exhaust air be discharged to the outside without being recirculated to other rooms.

e. An autoclave to decontaminate infectious waste is available in the same building with the animal facility. (See Appendix 9)

Animal Biosafety Level 3

1. Standard Practices

a. Doors to animal rooms are self-closing and self-locking and are kept closed when experiments are in progress.

b. Work surfaces are decontaminated following use or spills of viable materials. (See Section IV)

c. Eating, drinking smoking, and storing of food are not permitted in the animal room.

d. Personnel shall wash their hands after handling viable cultures and animals and before leaving the laboratory.

e. All procedures are carefully conducted to minimize the creation of aerosols.

f. An insect and rodent control program is in effect.

2. Special Practices

a. Cages are autoclaved before bedding is removed and before they are cleaned and washed.

b. Warp-around or solid-front gowns or uniforms are worn by personnel entering the animal room. Front-button laboratory coats are unsuitable. Protective gowns must remain in the animal room and must be decontaminated before being laundered.

c. Surgical-type masks or other respiratory protection devices are worn by personnel entering rooms housing animals infected with agents assigned to BL 3.

d. Access to the animal room is restricted by the supervisor or other responsible person to personnel who have been advised of the potential hazard and who need to enter on a program or service basis when experiments are in progress In general, persons who may be at increased risk of acquiring infection or for whom infection might be unusually hazardous are not allowed in the animal room. Persons at increased risk may include children, pregnant women, and individuals who are immunodeficient or immunosuppressed. The lab supervisor or other responsible person has the final responsibility for assessing individual circumstances and determining who may enter or work in the animal room.

e. The laboratory supervisor or other responsible person will assure that only persons who have been advised of the potential hazard and meet any specific requirements (e.g. immunization) may enter the animal room.

f. Hazard warning signs, incorporating the universal biohazard warning symbol are posted on access doors to animal rooms when materials containing, or animals infected with, agents assigned to BL 2 or higher are present. The hazard warning sign should identify the agent(s) in use, list the name of the animal room supervisor or other responsible person (s) and indicate any special conditions of entry into the animal room (e.g. immunizations, respirators).

g. Gloves are worn by personnel when handling infectious agents and animals. Gloves should be removed aseptically and autoclaved with other animal room wastes before being disposed of or reused.

h. All wastes from the animal room are autoclaved before being disposed of. All animal carcasses are disposed of as biohazardous waste.

i. The use of hypodermic needles and syringes is restricted to gavage, parenteral injection or aspiration of fluids from laboratory animals and diaphragm bottles. Serial dilutions of infected agents should not be done in diaphragm bottles with needles and syringes because of the hazards of autoinoculation and of aerosol exposure. Cannulas should be used instead of sharp needles whenever possible.

j. If floor drains are provided, the drain trap is always filled with water.

k. If vacuum lines are provided, they shall be protected with HEPA filters and liquid traps. (See Appendix 6)

l. Boots, shoe covers or other protective footwear and disinfectant foot baths are provided and used when indicated.

3. Biosafety Equipment

a. Personal protective clothing and equipment or other physical containment devices are used for all procedures and manipulations of infectious materials or infected animals.

b. Infected laboratory animals are housed in partial containment caging systems such as open cages place in ventilated enclosures, solid wall and bottom cages covered by filter bonnets or other equivalent primary containment systems.

4. Animal Facilities

a. The animal facility should be designed and constructed to facilitate cleaning and housekeeping and should be separated from areas which are open to unrestricted personnel traffic within the building. Separation shall be provided by an airlock or other access device which requires passage through two sets of doors to gain access to the animal room by a double door change room and shower.

b. A foot- or elbow-operated hand washing sink is provided near each animal room exit door.

c. Windows in the animal room are closed and sealed.

d. The animal room is provided with a ventilation system which creates an inward directional flow of air and assures that exhaust air is discharged directly to the outside or through the building exhaust system without being recirculated to any other area of the facility. Air within the animal room may be recirculated.

e. An autoclave for decontamination of wastes is available within the animal room or animal facility. Materials to be autoclaved outside the animal room are transported in a covered leakproof container. (See Appendix 9)

f. The surface, of walls, floors and ceilings are water resistant and easily cleaned. Openings in these surfaces are sealed or capable of being sealed to facilitate fumigation or space decontamination.

g. In animal facilities which have supply air systems, the supply air and exhaust air systems are electrically or mechanically interlocked to assure inward air-flow at all times.

h. The exhaust air from Class I or Class II biological safety cabinets can only be recirculated within the animal room after appropriate filtration through tested and certified cabinet exhaust HEPA filters.

i. Biological safety cabinets must be certified yearly (See Appendix 6).

F. Recommended Biosafety Levels for Infectious Agents and Infected Animals

The selection of an appropriate biosafety level for work with a particular agent is dependent upon a number of factors.

The most important of these include: the virulence, pathogenicity, biological stability, and communicability of the agent; the nature or functions of the laboratory; the quantity and concentration of the agent; the endemicity of the agent; and the availability of effective vaccines or therapeutic measures.

If a combination of increasingly stringent primary and secondary containment procedures and facilities are used, laboratory studies and manipulations can be safely conducted on agents that are correspondingly more hazardous.

In general, the biosafety level used for activities using infectious agents or infected animals should be commensurate with that required for the agent of highest virulence known or likely to be encountered in the course of contemplated work.

For example: all diagnostic sera of human origin should be considered potentially infectious for hepatitis and handled under conditions which reasonably preclude cutaneous, oral, and parenteral exposure to personnel: sputa should be considered as potentially infectious for tuberculosis and in addition should be handled under conditions which reasonably preclude the generation of aerosols. If in the course of diagnostic or other laboratory examinations there is evidence that the materials being studied contain only an agent of higher or lower risk than expected, the biosafety level should be raised or lowered accordingly.

In selecting the appropriate biosafety level, the laboratory manager and supervisor should determine the nature of the specific laboratory activity in which the infectious agent will be used.

Occasions will arise when the laboratory supervisor should select a biosafety level higher than that recommended in these guidelines.

For example, a higher biosafety level may be indicated by the unique nature of the proposed activity (e.g. the need for special containment of experimentally generated aerosols during inhalation studies) or by the proximity of the laboratory to areas of special concern (e.g. a laboratory located near patient care areas). Additionally, if the procedures involve large quantities or highly concentrated preparations of infectious agents or manipulations which are likely to produce aerosols, a higher biosafety level may be recommended.

The American Committee on Arthropod-Borne Viruses (ACAV) registered 424 arboviruses as of December 31, 1979. The ACAV Subcommittee on Arbovirus Laboratory Safety (SALS) publication Laboratory Safety for Arboviruses and Certain Other Viruses of Vertebrates (in press) has assigned these 424 agents to biosafety levels 2-4 based on risk assessments. Those 94 agents assigned to biosafety levels 3-4 and seven arboviruses which are indigenous or are commonly used in laboratories and assigned to biosafety level 2 are included in Section A. The remaining 330 arboviruses registered with ACAV as of December 31, 1979 are assigned to biosafety level 2 and are listed in Section B. Arboviruses not included on the December 31, 1979 registry will be individually assessed by SALS and subsequently assigned to an appropriate biosafety level.

IV. LABORATORY SPILLS

A problem that may occur in the laboratory is an overt biological spill. A spill that takes place in the open laboratory may create a serious problem. Every effort should be taken to avoid such occurrences. A spill poses less of a problem if it happens in a biological safety cabinet, provided splattering to the outside of the cabinet does not occur. Direct application of concentrated liquid disinfectant and a thorough wipe down of the internal surfaces of such cabinetry will usually be effective for decontaminating the work zone, but gaseous sterilants will be required to disinfect the interior sections of the cabinet. Each researcher must realize that in the event of an overt accident, research materials such as tissue cultures, media, and animals within such cabinets may well be lost to the experiment.

A. Spill in the Open Laboratory

Advance preparation for spill management is essential. A "spill kit" including leak proof containers, forceps, paper towels, sponges, disinfectant, respirators, and rubber gloves should be readily available. If potentially hazardous biological material is spilled in the laboratory, the first step is to avoid inhaling any airborne material by holding the breath and leaving the laboratory. Warn others in the area and go directly to the wash or change room area. If clothing is known or suspected to be contaminated, remove the clothing with care, folding the contaminated area inward. Discard the clothing into a bag or place the clothing directly in an autoclave. Wash all potentially contaminated body areas as well as the arms, face, and hands. Shower if facilities are available. Reentry into the laboratory should be delayed for a period of thirty minutes to allow the dissipation of the aerosol generated by the spill.

Protective clothing should be worn when entering the laboratory to clean the spill area. Rubber gloves, autoclavable footwear, an outer garment, and a respirator should be worn. If the spill was on the floor, do not use a surgical gown that may trail on the floor when bending down. Take the " spill kit" into the laboratory room, place a discard container near the spill, transfer large fragments of material into it and replace the cover. Using a hypochlorite containing solution (1000 ppm available chlorine), iodopher solution containing 1600 ppm iodine or other appropriate disinfectant, carefully pour the disinfectant around and into the visible spill. Avoid splashing. Allow 15 minutes contact time. Use paper or cloth towels to wipe up the disinfectant and spill, working toward the center of the spill. Discard towels into a

discard container as they are used.
Wipe the outside of the discard containers, especially the bottom, with a towel soaked in a disinfectant. Place the discard container and other materials in an autoclave and sterilize.

Remove shoes, outer clothing, respirator and gloves and sterilize by autoclaving or exposure to ethylene oxide. Wash hands, arms and face, or if possible, shower.

B. Spill in a Biological Safety Cabinet

A spill that is confined to the interior of the biological safety cabinet should present little or no hazard to personnel in the area. However, chemical disinfection procedures should be initiated at once while the cabinet ventilation system continues to operate to prevent escape of contaminants from the cabinet. Spray or wipe walls, work surfaces and equipment with a disinfectant. A disinfectant with a detergent has the advantage of detergent activity which will help clean the surfaces by removing both dirt and microorganisms. A suitable disinfectant is a 3% solution of an iodopher such as Wescondyne or a 1:100 dilution of house-hold bleach (e.g. Chlorox) with 0.7% nonionic detergent. The operator should wear gloves during this procedure. Use sufficient disinfectant solution to ensure that the drain pans and catch basins below the work surface contain the disinfectant. Lift the front exhaust grill and tray and wipe all surfaces. Wipe the catch basin and drain the disinfectant into a container. The disinfectant, gloves, wiping cloth and sponges should be discarded into an autoclave pan and autoclaved.

The above procedure will not disinfect the filters, blower, air ducts or other interior parts of the cabinet. If the entire interior of the cabinet needs to be sterilized, contact EH&S.

C. Biological Spill Response Guidelines

These guidelines are intended to assist the principal investigator, laboratory supervisor, and other responsible individuals who may be involved in the cleanup of biological spills. This guide outlines the basic procedures for dealing with some of the biological spills that may be encountered in a research laboratory. All lab personnel should refer to the specific spill response procedures before initiating their experiments.

Biosafety Level 1 (BL1) Spill

- Notify others in the area, to prevent contamination of additional personnel and environment.
- When BL1 spills occur outside the lab (e.g. hallways, common rooms & corridors) report these BL-1 spills to: (**1) Lab Director (2) Biosafety Officer (620-2019)**
- Remove any contaminated clothing and wash exposed skin with soap and water.

Clean-up of BL1 Spill

- Wearing gloves and lab coat, cover spill with paper towels, pour disinfectant around the spill allowing it to mix with spilled material. Allow suitable contact time, at least 15 min.
- Pick up any pieces of broken glass with forceps and place in a sharps container.
- Discard all disposable materials used to clean up the spill into a biohazard bag.
- Wash hands with soap and water.

Biosafety Level 2 (BL2) Spill

- Notify others in the laboratory regarding the spill
- Close door, and post with a warning sign.
- Remove contaminated clothing, turning exposed areas inward, and place in a biohazard bag.

- Wash all exposed skin with soap and water.
- Inform Lab director, University Police Department (911), and Biosafety Officer (620-2019)

Clean-up of BL2 Spill

- Allow aerosols to disperse and or settle for at least 30 minutes before reentering the laboratory (if spill outside cabinet). Assemble clean-up materials (disinfectant, paper towels, biohazard bags, and forceps).
- Put on protective clothing (lab coat, facemasks/face protection, utility gloves, and booties if necessary).
- Cover the area with disinfectant-soaked towels, and then carefully pour disinfectant around the spill. Avoid enlarging the contaminated area. Use more concentrated disinfectant as it is diluted by the spill. Allow at least a 20 minute contact time.
- Pick up any sharp objects with forceps and discard in a sharps container.
- Soak up the disinfectant and spill using mechanical means, such as an autoclavable broom and dustpan, since there may be sharps under the paper towels, and place the materials into a sharps container.
- Smaller pieces of glass may be collected with cotton or paper towels held with forceps. If no sharps were involved in the spill discard the materials into an autoclave bag.
- Wipe surrounding areas (where the spill may have splashed) with disinfectant.
- Spray the area with 10% household bleach solution and allow to air-dry (or wipe down with disinfectant-soaked towels after a 20-minute contact time).
- Place all contaminated paper towels and any contaminated protective clothing into a biohazard bag and autoclave.
- Wash hands and exposed skin areas with soap and water.

D. Engineering Controls

Engineering controls are tools or equipment that, when used properly, provide significant protection to the operator as well as other laboratory occupants. Examples include biological safety cabinets, autoclaves, and sharps containers. As is true of most tools, there are correct (proper) and improper ways to use engineering controls. Given that there are considerable adverse consequences if used improperly, correct usage of engineering controls is critical. When used properly, they have proven to be, in most circumstances, the most effective and practical way to achieve a safety goal.

A. Biological Safety Cabinets (BSCs)

BSCs are designed to provide personnel, environmental, and product protection when appropriate use/procedures are followed. They are among the most effective and commonly used primary containment devices in laboratories working with infectious agents. Three types of BSCs (Class I, II, and III) have been developed to meet various research and clinical needs. BSCs use HEPA (high efficiency particulate air) filters in their exhaust and/or supply systems. Biological safety cabinets must not be confused with other laminar flow devices or "clean benches." Horizontal flow cabinets that direct air toward the operator **should never be used for handling infectious, toxic or sensitizing materials**. Class I and II biosafety cabinets, when used in conjunction with good microbiological techniques, provide an effective partial containment system for safe manipulation of moderate and high-risk microorganisms, (i.e. BL 2 and 3 agents).

• **Class I Biological Safety Cabinet** is a ventilated cabinet for personnel protection with an unrecirculated inward airflow away from the operator. This unit is fitted with a HEPA filter to protect the environment from discharged agents. A Class I BSC is suitable for work involving low to moderate risk agents, where there is a need for containment, but not for product protection.

• **Class II Biological Safety Cabinet** is a ventilated cabinet for personnel, product and environmental protection, which provides inward airflow and HEPA-filtered supply and exhaust air. There are three basic types of Class II BSCs: Type A, Type B, and 100% exhaust. Type B cabinets are further sub-typed into types B1, B2, and B3. The major differences between the three types is the percent of air that is exhausted or recirculated, and the manner in which exhaust air is removed from the work area.

• **Class III Biological Safety Cabinet or glove box** is a totally enclosed ventilated cabinet, which provides the highest attainable level of protection to personnel, environment and product. The supply air is HEPA-filtered and exhaust air has two HEPA-filters in series. Work is performed in the cabinet through glove ports with O-ring for attaching arm-length gloves to cabinet.

It is important to note that laminar flow clean benches must not be utilized for work with biohazardous or chemically hazardous agents.

Table 1 – Selection of a Biological Safety Cabinet through Risk Assessment.

Biological Risk Assessed	Protection Provided			BSC Class
	Personnel	Product	Environmental	
BSL 1-3	YES	NO	YES	I
BSL 1-3	YES	YES	YES	II (A, B1, B2, B3)
BSL 4	YES	YES	YES	III B1, B2

Clean benches provide product protection by ensuring that the product is exposed only to HEPA-filtered air. They do not provide protection to personnel or the ambient environment. The correct location, installation, and certification of a biological safety cabinet are critical to its performance in containing infectious aerosols. All BSCs used for must be inspected and certified annually. Inspection and re-certification is mandatory if the cabinet is relocated, experiences major repairs or after a filter change etc.

B. Safe and Effective Use of Biosafety Cabinets

• All personnel must b trained prior to using the BSC.

• Equipment and/or supplies should not be stored inside the cabinet; unnecessary objects can disrupt airflow in the cabinet.

• Do not use the top of the cabinet for storage. The HEPA filter can be damaged, disrupting the balance of airflow.

• Do not place any objects that will block either the front air intake grille, or block the rear exhaust grille.

• Close the sash when the cabinet is not in use. Keeping the sash closed between uses will assist in minimizing cabinet contamination. Always leave the BSC running.

• Do not eat, drink, chew gum, or store food near the cabinet.

• Avoid sudden movement in or out of the cabinet, as well as areas directly adjacent to the cabinet. Sudden movements can disrupt the airflow and compromise safety. Move arms slowly when removing or introducing new items into the BSC.

• Segregate contaminated and clean items. Work from "clean to dirty"
• Protect the building vacuum system from biohazards by placing a cartridge filter between the vacuum trap and the source valve in the cabinet.

• Always decontaminate the interior surfaces prior to working in the cabinet. Clean up spills in the cabinet immediately.

• When work is finished, remove all materials and wipe all interior surfaces with 70% alcohol or any other disinfectant suitable for the agent(s) in use.

• Always extinguish and remove the Bunsen burner from the hood prior to decontamination with alcohol.

• Remove lab coat, gloves and other personal protective equipment (PPE) and wash hands thoroughly before leaving the laboratory.

Table 2 – Summary of Recommended Biosafety Levels for Infectious Agents

BSL	Agents	Practices	Safety Equipment (Primary Barriers)	Facilities (Secondary Barriers)
1	Not known to consistently cause disease	Standard Microbiological Practices	None required	Open bench top sink required

	in healthy adults			
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

V. Biohazardous Waste Disposal

All biohazardous waste is required to be inactivated prior to disposal. The preferred method of inactivation is steam sterilization (autoclaving) although chemical inactivation may be appropriate in some cases. Liquid waste should be poured down the drain (sanitary sewer) following chemical inactivation.

BL 2 agents, veterinary pathogens, recombinant DNA materials and plant pathogens regulated by APHIS and DPI must be inactivated prior to disposal. Materials that are considered biohazardous waste by state law include, but are not limited to, the following: nonliquid human tissue and body parts, human "disease-causing agents", discarded sharps, human blood, human blood products and human body fluids. Other materials that are covered under this rule are bandages, gauze or other materials that are saturated with human blood or body fluids, medical tubing and other devices with visible blood adhering to them and any contaminated material that represents a "significant risk of infection".

The local landfill does not accept "red-bagged" material or any "medical waste", even if inactivated. This material must be inactivated and collected by a licensed biomedical waste vendor for appropriate disposal.

If the transport of contaminated waste outside of the laboratory becomes necessary (to an autoclave or medical waste receptacle) it shall be in closed, non-porous containers.

Sharp objects must be placed in puncture-proof containers. Material to be transported must be identified as biohazardous either by red-bagging or by labeling with the universal biohazard symbol. The date and name of the lab supervisor or principal investigator should be clearly visible.

The storage of contaminated waste is restricted to within the generating laboratory. Biohazardous waste may not be stored for longer than 24 hours without being inactivated. Following inactivation, biohazardous waste may not be stored longer than 36 hours prior to collection.

Needles, scalpels and razor blades must be placed in labeled, puncture-proof "sharps" containers. Sharps that are contaminated require steam sterilization or chemical inactivation prior to disposal.

Please note that it is the responsibility of the principal investigator or the laboratory supervisor or lab manager to ensure compliance with this policy.

VI. Medical Surveillance and Accident Reporting

A. Medical Surveillance

The purpose of a medical surveillance program is to insure that employees are physically fit for employment, properly protected against potentially harmful exposures and appropriately diagnosed should illness or exposure occur.

It is the responsibility of the principal investigator, laboratory supervisor or laboratory manager with the assistance of EH&S to consider the potential hazards associated with each position and provide the examining physician with guidelines for the conduct of the medical exam.

Please be sure to consult the agent listing (V, D) and the section on immunoprophylaxis (Appendix 5) of vaccination requirements when considering medial examination. Note that all individuals working with BL 3 agents are required to have a serum sample stored for baseline information. In addition, note that certain classes of individuals are at increased risk of infection. This would include those on long term antibiotic treatments, the immunosuppressed, pregnant employees, and individuals receiving chemotherapeutic agents. It is the responsibility of the employee to call any of the above conditions to the attention of his or her supervisor so that proper safety precautions can be implemented.

B. Accident Reporting

All accidents involving biological agents should be reported to the lab supervisor, lab manager and EH&S. This includes, but is not be limited to, animal bites, accidental exposure to infectious agents and spills of potentially hazardous agents. Please utilize the UNF Accident Reporting Form (Appendix 10) to report these occurrences.

VII. Registration of Experiments

Projects that require registration with EH&S.

A. Bio-agents:

1. Human, animal, or plant pathogens that are BL 2 and above. (See Section V.) Agents that are regulated by government agencies (USDA/APHIS, DPI, etc.).

Unknown human and/or animal pathogens (considered BL 2 until identified). These include bacteria, viruses, fungi, mycoplasmas, parasites, infected animals and/or tissues.

2. Cell cultures:

a. All cells that have been immortalized with EBV or a retrovirus,

b. All tumorigenic primate and human cell lines,

c. All primary human tumor lines.

B. Recombinant DNA projects:

1. All recombinant DNA projects including the growth of bacteria for probe (plasmid or phage preparation). Projects must be registered regardless of where the material came from or who originally constructed it.

2. The use of transgenic animals and plants, regardless of where the materials came from or who originally constructed/ regenerated them.

C. Acute toxins:

1. The use and storage of chemicals with a mammalian LD_{5O} of < 100 g/kg.

VIII. Aquisition , Possession and Shipment of Biologicals

The acquisition, possession or shipment of certain organisms pathogenic for either man, animal or plant is regulated by various state and federal agencies. General information is provided below.

A. Human Pathogens and Related Materials

The importation or subsequent receipt of etiologic agents and vectors of human disease is subject to Foreign Quarantine Regulations (42 CFR, Section 71.156). Permits authorizing the importation or receipt of regulated materials and specifying conditions under which the agent or vector is shipped, handled, and used are issued by the Centers for Disease Control.

The interstate shipment of indigenous etiologic agents, diagnostic specimens and biological products is subject to applicable packaging, labeling and shipping requirements of the Interstate Shipment of Etiologic Agents (42 CFR Part 72). Packaging and labeling requirements for interstate shipment of etiologic agents are summarized and illustrated in Appendix 2.

Additional information on the importation and interstate shipment of etiologic agents of human disease and other related materials may be obtained by contacting EH&S or:

Office of Biosafety
Centers for Disease Control
1600 Clifton Road, NE
Atlanta, Georgia 30333
Telephone: (404) 329-3883

B. Animal Pathogens

Non-indigenous pathogens of domestic livestock and poultry may require special laboratory design, operation, and containment features not generally addressed in this manual. The importation, possession, or use of the following agents is prohibited or restricted by law or by U. S. Department of Agriculture regulations or administrative policies:

African horse sickness virus
African swine fever virus
Besnoitia besnoiti
Borna disease virus
Bovine infectious petechial (Gamjam virus)
Newcastle Disease virus
Mycoplasma mycoides
Mycoplasma agalactiae
Pseudomonas mallei
Rickettsia ruminantium
Rift Valley fever virus
Rinderpest virus
Sheep pox virus
Swine vesicular disease virus
Foot and mouth disease virus
Fowl plague virus
Histoplasma (Zymonema) farciminosum
Louping ill virus
Lumpy skin disease virus
Nairobi sheep disease virus
Camel pox virus
Ephemeral fever virus
Teschen disease virus
Trypanosoma vivax
Trypansoma evansi
Trypanosoma parva
Theilaria annulata
Theileria lawrencei
Thaileria bovis
Theileria hirce
Vesicular exanthema virus
Wesselsbron disease virus

Hog cholera virus

The importation, possession, use or interstate shipment of animal pathogens other than those listed above may also be subject to regulations of the U. S. Department of Agriculture.

Additional information may be obtained by writing directly to:

Chief Staff Veterinarian
Organisms and Vectors; Veterinary Services
Animal and Plant Health Inspection Services
U. S. Department of Agriculture
Hyattsville, Maryland 20782
Telephone: (301) 436-8017

C. Plant Pathogens

The introduction of serious plant pathogens has been of long- standing concern to Florida agriculture. The number of plant pathogens and their vectors introduce into the United States increases steadily each year. These present an ever present threat to agriculture.

Three federal statutes--the Animal Quarantine Act of 1903 = Plant Quarantine Act of 1912, and the Federal Plant Pest Act of 1957--and related state laws prohibit the importation and movement of plant pathogenic organisms, their vectors, and any articles that might harbor these organisms, except by occasional special permits.

The introduction and dissemination of all hazardous or potentially hazardous plant pathogens or articles that may harbor these organisms into or within the State of Florida must be regulated or prohibited to ensure the protection of agriculture.

Any plant pathogenic organism in any living stage capable of reproduction that is requested for movement into or within the State of Florida for research studies or any purpose must be approved by the Division of Plant Industry, Florida Department of Agriculture and Consumer Services (Chapter 581.083, Florida Statutes). The approval by the Division is based on recommendations from the Florida Plant Pathogen Introduction Committee and other pertinent reference sources the Division may choose to consult.

General agricultural plant pathogens include:

Algae (parasitic or any parasitc plant)	Rickettsia	Viroids
Bacteria	Viruses	Fungi
Nematodes	Mycoplasmas	

And any others to be designated

Any person wishing to introduce a plant pathogen into the State of Florida should make application on USDA PPQ Form 526 to:

Director
Division of Plant Industry
P.0. Box 1269
Gainesville, Florida 32602

Upon approval by the Division, the request will be referred to the USDA Plant Protection and Quarantine Division, Hyattsville, Maryland 20782, which will issue a shipping permit to the applicant, if the request also meets with their approval. The shipping permit must be attached to the package containing the plant pest.

Anyone wishing to send a plant pest (culture or other living stage) into another state should contact the regulatory official of that state's department of agriculture concerning the request. The name and address of the regulatory official of any state will be furnished upon request.

If necessary, certain safety procedures will be established for the investigator to follow and will be periodically monitored by state and federal regulatory officials.

If you have specific questions concerning the movement of plant pathogens interstate or internationally, you may wish to consult the following:

Staff Officer
Regulatory Support
Plant Protection and Quarantine
USDA - APHIS
Room 635, Federal Building
Hyattsville, Maryland 20782

District Director
Plant Protection & Quarantine
USDA - APHIS
Room 1524, Federal Building
51 SW First Avenue
Miami, Florida 33130

Appendix

UNIVERSITY OF NORTH FLORIDA
Registration of Biological Agent

Lab Supervisor/Principal Investigator:_____

Title _____ Project Bldg:_____

Department:_____ Project Rm# :_____

Telephone No:_____ Grant # : _____

Grant/Project Title:_____

_____

1.Description:

(a) Name of Organism:_____ (b) assigned biosafety level:_____

(c) Self-sufficient description of the experiment:_____

_____

_____

2. Information on Safety Procedures:

(a) Personal Protection: _____

_____

(b) Waste disposal:_____

_____

3. The undersigned individuals will be involved in the above-mentioned experimentation. They are familiar with, and agree to abide by the current University of North Florida guidelines.

_____ _____

_____ _____

_____ _____

I attest to the fact that the above named individuals are properly trained in this area of experimentation.

_____

Lab Supervisor/ Principal Investigator Date

4. The indicated sites have been inspected and are in compliance with the University of North Florida guidelines.

_____

Environmental Health & Safety Officer Date

REGISTRATION OF EXPERIMENTS
(Projects That Require Registration With EH&S)

A. Biological Agents

1. Human, animal or plant pathogens that are *BL 2) and above (i.e., Escherichia coli non K-12 strain, Herpesvirus, Vaccinia, etc.).

2. Agents that are regulated by government agencies (USDA/APHIS, DPI, etc.) such as Histoplasma faciminosun, Pseudomonas mallei, etc.

3. Unknown human and/or animal pathogens (considered BL 2 unidentified). These include bacteria, fungi, parasites, viruses, mycoplasmas, and infested animals and/or tissues.

4. Cell Cultures

a. All cells that have been immortalized with EBV or a retrovirus.

b. All tumorigenic primate and human cell lines.

c. All primary human tumor lines.

B. Recombinant DNA Projects

1. ALL recombinant DNA projects including the growth of bacteria for probe (plasmid or phage preparation). Projects must be registered regardless of where the material came from or who originally constructed it.

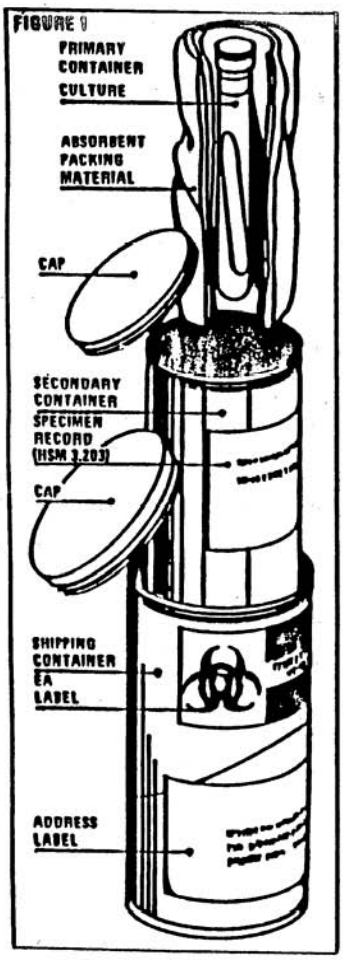
2. The use of transgenic animals and plants, regardless of where the materials came from or who roiginally constructed/regenerated them.

C. Acute Toxins

The use and storage of chemicals with a mammalian LD_{50} of $\leq$ 100 ug/kg (i.e., Clostridium botulinum, dioxin, etc.).

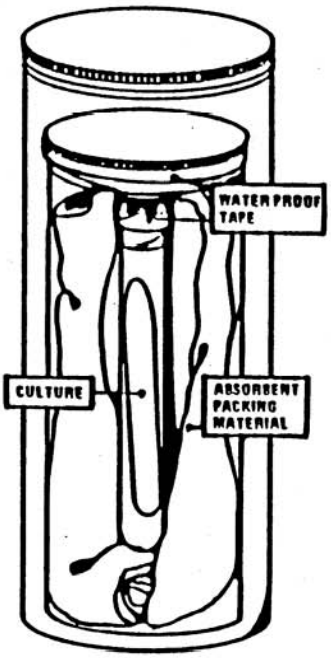
Appendix 2

Packaging and Labeling of Etiologic Agents



PACKAGING AND LABELING OF ETIOLOGICAL AGENTS

FIGURE 2



CROSS SECTION OF PROPER PACKING

The Interstate Shipment of Etiologic Agents (42 CFR, Part 72) was revised July 21, 1980 to provide for packaging and labeling requirements for etiologic agents and certain other materials shipped in Interstate traffic.

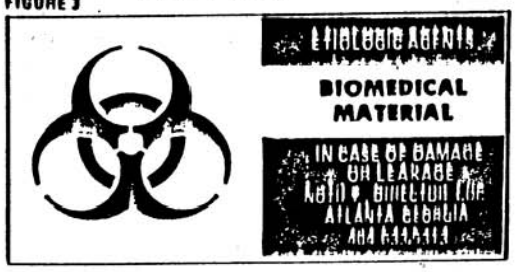
Figures 1 and 2 diagram the packaging and labeling of etiologic agents in volumes of less than 50 ml. in accordance with the provisions of subparagraph 72.3 (a) of the cited regulation. Figure illustrates the color and size of the label, described in subparagraph 72.3 (d)(1 - 5) of the regulations, which shall be affixed to all shipments of etiologic agents.

For further information on any provision of this regulation contact:

Centers for Disease Control
Attn: Biohazards Control Office
1600 Clifton Road
Atlanta, Georgia 30333

Telephone: 404-329-3883
FTS-236-3883

FIGURE 3 (Red on White)



Appendix 3

PLANT PATHOGEN INFORMATIONAL FORM

Florida Department of Agriculture & Consumer Services
Division of Plant Industry

(This form must accompany "Application to Move Live Plant Pests into Florida-PPQ Form 526")

1. State purpose and justification for request. Indicate why indigenous pathogens would not serve purpose of investigation.

2. List all personnel who would be involved with project.

3. Indicate location of work and briefly describe test facility and equipment (special if applicable) to be used.

4. Time required for completion of project.

5. Briefly describe research methods to be used during investigation.

6. Indicate sanitation procedures to be used to contain pathogen in testing area and security procedures to be used to prohibit unauthorized personnel from entering investigation area.

7. List procedures to be used at conclusion of tests to (a) destroy pathogen, host plants and vectors, and (b) to clean the testing facility.

Appendix 4

IMMUNOPROPHYLAXIS

Vaccines for which benefits (levels of antibody considered to be protective) clearly exceed the risks (local or systemic reactions) should be required for all clearly identified at-risk personnel. Examples of such preparations include vaccines against yellow fever, rabies and poliomyelitis. This type of vaccination should be made a prerequisite to employment.

If necessary, pre-employment physicals are provided to new employees at the request of the prospective employer. The lab supervisor or lab manager should make this request to EH&S and Human Resources.

Recommendations for giving less efficacious vaccines (i.e. those associated with high rates of local or systemic reactions or those that produce increasingly severe reactions with repeated use) should be carefully considered. Products with these characteristics (e.g. cholera vaccine, tularemia vaccine, typhoid vaccine) should be recommended but may not ordinarily be required for employment. A complete record of vaccines received on the basis of occupational requirements or recommendations should be maintained in the employee's permanent medical file.

The following table summarizes vaccination practices at the U.S. Government Centers for Disease Control. These practices were adapted from the Recommendations of the Public Health Service Advisory Committee on Immunization Practices for specific application to at-risk personnel working in or entering laboratory areas. Additional vaccination recommendations may be found in the agent listings, section V, D.

Appendix 5

RECOMMENDATIONS FOR PROPHYLACTIC IMMUNIZATION OF LABORATORY PERSONNEL WORKING WITH INFECTIOUS AGENTS[1]

VACCINE OR TOXOID	PERSONNEL WORKING DIRECTLY WITH THE AGENT OR WITH INFECTED ANIMALS	PERSONNEL WORKING WITH OR ENTERING ROOMS WHERE AGENT OR ANIMALS ARE PRESENT
Anthrax Vaccine	R[2]	R[3]
Botulism Toxoid (Pentavalent)[4]	R	NR
Cholera Vaccine	R	NR
Diphtheria-Tetanus Toxoids	R	NR
Eastern Equine Encephalitis Vaccine[4]	R	R
Influenza Vaccine[5]	NR	NR
Measles Vaccines (Rubella and Rubeola)	R	NR
Mumps Vaccine	NR	NR
Poliomyelitis Vaccine[5]	R	R
Q Fever Vaccine[4]	R	R
Rabies Vaccine	R	R
Rift Valley Fever Vaccine[4]	R	R
Rocky Mountain Spotted Fever Vaccine[6]	R	R
Rubella Vaccine	R	NR
Russian Spring-Summer Encephalitis Vaccine[4]	R	R
Smallpox Vaccine	R	R
Tularemia Vaccine[4]	R	R
Typhoid Vaccine	R	R
Typhus Vaccine[6]	R	R
Venezuelan Equine Encephalitis Vaccine[4]	R	R
Western Equine Encephalitis Vaccine[4]	R	R
Yellow Fever Vaccine	R	R

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[1] Adapted from Recommendations of the PHS Advisory Committee on Immunization Practices and Lab Safety at the Center of Disease Control

[2] R = Required

[3] NR = Not Required

[4] Unlicensed Biological Product. Available under IND or other limited use protocol.

[5] Unvaccinated or serologically negative personnel should be vaccinated.

[6] Commercially licensed vaccine not currently available.

Appendix 6

BIOLOGICAL SAFETY CABINETS

The Class I biological safety cabinet is an open-fronted, negative- pressure, ventilated cabinet with a minimum inward face velocity at the work opening of 75 feet per minute(0.4 m/s). The exhaust air from the cabinet is filtered by a high efficiency particulate air (HEPA) filter This cabinet may be used in three operational modes: with a full-width open front, with an installed front closure panel not equipped with gloves and with an installed front closure panel equipped with arm-length rubber gloves.

The Class II vertical laminar-flow biological cabinet is an open- fronted, ventilated cabinet with an average inward face velocity at the work opening of 75 feet per minute (0.4 m/s). This cabinet provides a HEPA- filtered recirculated mass airflow within the work space. The exhaust air from the cabinet is also filtered by HEPA filters.

The Class III cabinet is a totally enclosed ventilated cabinet of gas-tight construction. Operations within the Class III cabinet are conducted through attached rubber gloves. When in use, the Class II cabinet is maintained under negative air pressure of at least 0. 5 inches water (124.5 N/m2 or 0.9 mm Hg). Supply air is drawn into the cabinet through HEPA filters. The cabinet exhaust air is filtered by two HEPA filters installed in series. The exhaust fan for the Class III cabinet is generally separate from the exhaust fans of the facility' s ventilation system.

Personnel protection provided by Class I and Class II cabinet is dependent on the inward airflow. Since the face velocities are similar, they generally provide an equivalent level of personnel protection. The use of these cabinets alone, however, is not appropriate for containment of highest-risk infectious agents because aerosols may accidentally escape through the open front.

The use of a Class II cabinet in the microbiological laboratory offers the additional capability and advantage of protecting materials contained within it from extraneous airborne contaminants. This capability is provided by the HEPA-filtered, recirculated mass airflow within the work space.

The Class III cabinet provides the highest level of personnel and product protection. This protection is provided by the physical isolation of the space in which the infectious agent is maintained. When these cabinets are required, all procedures involving infectious agents are contained within them. Several Class III cabinets are therefore typically set up as an interconnected system. All equipment required by the laboratory activity, such as incubators, refrigerators and centrifuges, must be an integral part of the cabinet system.

Double-doored autoclaves and chemical dunk tanks are also attached to the cabinet system to allow safe introduction and removal of supplies and equipment.

Note: All Class III cabinets and those Class II cabinets being used with category 3 agents are required to be tested and certified yearly. Contact Environmental Health and Safety for arrangements. All biological safety cabinets are required to be tested and certified prior to initial use and following relocation.

Appendix 7

VACUUM SYSTEM FILTRATION

The aspiration of tissue culture media from monolayer cultures and of supernatants from centrifuged samples into primary collection flasks is a common laboratory procedure. Protection should be provided against pulling biohazardous aerosols or overflow fluid into the vacuum system. This protection is provided by the use of an air filter in the line immediately leading into the house vacuum line and an overflow flask for liquids between the collection flask and the air filter.

Two techniques for protecting the vacuum system are shown. A cartridge-type filter provides an effective barrier to passage of aerosols into the house vacuum system. The filter has a capacity to remove airborne particles 450 nm (0.45μ) or larger.

For assembling either apparatus, flexible tubing is used of appropriate inside diameter for the flask and filter fittings and of sufficient wall thickness for the applied vacuum. Filter flasks of capacities from 250 to 4000 ml may be used for the overflow flask, depending on available space and amount of fluid that could be accidently aspirated out of the collection flask.

The overflow flasks contain a disinfectant solution appropriate for the biohazardous material under study. It is essential that an antifoam, such as Dow Corning Antifoam A, be added to the overflow flask, since bubbling of air through the disinfectant probably will cause considerable foam which, if allowed to reach the filter, will shut off the vacuum.

If the filter becomes contaminated or requires changing, the filter and flask can be safely removed by clamping the line between filter and vacuum source. The filter and flask should be autoclaved before the filter is discarded. A new filter can then be installed and the assembly replaced.

The apparatus shown is composed of two suction flasks, a filter, rubber stoppers, flexible vacuum tubing, glass tubing and a small glass sparger. Various small fritted glass or ceramic spargers or gas dispersion tubas are commercially available. The coarse or medium porosity sparger assures that any aerosol passing through the collection flask is dispersed in small bubbles so that adequate contact is made with the disinfectant solution.

The apparatus depicted in B has the feature of automatically shutting off the vacuum when the storage flask is full. It consists of a 1 liter filter flask with a small glass Buchner funnel (15 ml capacity, 29 mm filter disc) inserted upside down in a No. 8 rubber stopper in the mouth of the flask.
A hole, 2 cm in diameter, is cut into the bottom of the stopper with a cork borer and of sufficient depth that the filter disc is level with the bottom of the stopper.

A 14 gr (1/2 oz) rubber bulb measuring 6 cm (2 3/8 inches) in length and 3.2 cm (1 1/4 inches) in diameter, with the end plugged with a solid glass rod measuring 0.6 cm (1/4 inch) in diameter and approximately 5.7 cm (2 1/2 inches) in length, is placed inside the flask.

If liquids enter the overflow flask the rubber bulb rises until it presses against the mouth of the Buchner funnel and shuts off the vacuum. The entire unit is autoclavable, but the filter assembly should be thoroughly dried before reuse.

Appendix 8

CHEMICAL DISINFECTANTS

Following is a list of commonly used disinfectants. If a chemical disinfectant is required, please be sure to choose a compound which is active against the agent you are using. This list is not complete and you are urged to contact Environmental Health and Safety should additional information be required.

1. Chlorine

This halogen is a universal decontaminant active against all microorganisms, including bacterial spores. Chlorine combines with protein and rapidly decreases in concentration in its presence. Free, available chlorine is the active element. It is a strong oxidizing agent, corrosive to metals. Chlorine solutions will gradually lose strength so that fresh solutions must be prepared frequently. Sodium hypochlorite is usually used as a base for chlorine decontaminants. An excellent decontaminant can be prepared from household or laundry bleach. These bleaches, usually contain 5.25% available chlorine or 52,500 ppm. If diluted 1:100, the solution will contain 525 ppm of available chlorine and if a nonionic detergent such as Naccanol is added in a concentration of about 0.7% a very good decontaminant is created. These are recommended for certain disinfecting procedures provided the available chlorine needed is considered. Low concentrations of available chlorine (50 to 500 ppm) are active against vegetative bacteria and most viruses. For bacterial spores, concentrations of approximately 2500 ppm are needed.

2. Iodine

The characteristics of chlorine and iodine are similar. One of the most popular groups of decontaminants used in the oncologic laboratory is the iodophors and Wescodyne is perhaps the most popular. The range of dilution of Wescodyne recommended by the manufacturer is 1 oz. in 5 gal. (28.35gr. in 18.9 L) of water giving 75 ppm of available iodine, to 3 oz. in 5 gal. (85 gr. in 18.9 L) giving 75 ppm. At 75 ppm, the concentration of free iodine is 0.0075%. This small amount can be rapidly taken up by any extraneous protein present. Clean surfaces or clear water can be effectively treated by 75 ppm available iodine, but difficulties may be experienced if any appreciable amount of protein is present. For bacterial spores, a dilution of 1:40 is recommended by the manufacturer giving 750 ppm.

For washing the hands, it is recommended that Wescodyne be diluted 1:10 or 10% in 50% ethyl alcohol (a reasonably good decontaminant itself) which will give 1600 ppm of available iodine, at which concentrations relatively rapid inactivation of any and all microorganisms will occur. Iodophors have a built-in indicator. If the solution is brown or yellow, it is still active. Iodophors are relatively harmless to man. They can be readily inactivated and their stains can be removed easily with solutions of $Na_2S_2O_3$ (sodium thiosulfate).

3. Formaldehyde-Alcohol

Solutions of 8% formalin in 70% alcohol are considered very good for disinfection purposes because of their effectiveness against vegetative bacteria, spores and viruses. For many applications this is the disinfectant of choice. However, it should be stressed that formaldehyde is a known irritant, sensitizer and suspect carcinogen. Therefore, its use should be restricted to those applications which minimize exposure potential.

4. Alcohols

In concentrations of 70 to 95% alcoholic solutions are good general-use disinfectants, but they exhibit no activity against bacterial spores.

Appendix 9

AUTOCLAVE TESTING

Steam sterilizers are required to be tested by the user every six months. Any commercially available test indicator that uses bacterial spores (Bacillus stearothermophilus) is appropriate. A positive control (do not sterilize one vial) must be included as a part of each test. The dates of certification must be posted near the autoclave.

Autoclaves used with HUMAN pathogens or tissues (including blood) are required by state law to be tested after every 40 hours of use. Additionally, each load of material inactivated in that autoclave must be logged as follows: date, time, operator, amount of waste and confirmation of sterilization (autoclave tape).

Biohazard bags that are used for inactivation of human pathogens or tissues must meet impact requirements (165 grams) and tearing resistance (480 grams) requirements. Written documentation from the bag manufacturer must be kept on file.

Appendix 10

UNIVERSITY OF NORTH FLORIDA
ACCIDENT INVESTIGATION FORM
(Please Print)

Name: _____ N#:_____ DOB: _____
Gender: _____ Department: _____
Job Title: _____ Time in Job: _____
Unit: Auxiliary: _____ Contracts and Grants: _____ E&G:_____ Local Funds:____
Status: Faculty/Staff: _____ Student: _____ Visitor: _____

Description of Accident (Use Additional Sheets If Necessary):
_____
_____
_____
_____
_____
_____

Accident: Date: _____ Time: _____ (a.m./p.m.)
Location: Building:_____Room:_____ Other:_____

Accident Class: Fire: _____ Hazardous Material: _____ Injury: _____ Vehicle: _____ Other: _____

What Could Have Been Specifically Done To Prevent The Accident (Cannot Use "Be More Careful")?
_____
_____
_____
_____

Corrective Actions Recommended:
_____
_____
_____

Use Codes Provided On The Reverse Side Of This Form
To Complete This Section

Nature of Injury: _____ Part of Body Affected: _____ Source of Injury: _____
Incident Type: _____ Unsafe Condition: _____ Unsafe Act: _____
Cause: _____ Severity of Injury: _____ Date Investigated:_____
Investigated By: _____ Phone Extension: _____

FOR EH&S USE ONLY:
Recommendations Forwarded To: _____ Date: _____

INJURY CODES

NATURE OF INJURY *(Cut, Bruises, etc.)*

NJ-01 Abrasion, Scratch
NJ-02 Amputation
NJ-03 Bite, Stings
NJ-04 Burn
NJ-05 Burn, Electrical
NJ-06 Chemical Sensitivity-Acute
NJ-07 Chemical Sensitivity-Chronic
NJ-08 Contusion, Crushing Bone
NJ-09 Contusion
NJ-10 Cut, Laceration, Puncture
NJ-11 Dermatitis, Rash
NJ-12 Electric Shock, Electrocution
NJ-13 Eye, Flash Burn
NJ-14 Eye, Irritation
NJ-15 Fracture
NJ-16 Hearing Loss
NJ-17 Heat Exhaustion
NJ-18 Hernia, Rupture
NJ-19 Inflammation, Irritation
NJ-10 Multiple Injuries
NJ-21 Occupational Illness
NJ-22 Repetitive Motion
NJ-23 Respiratory
NJ-24 Sprains, Strains, Dislocation

INCIDENT TYPE *(Event Which Directly Resulted in the Injury)*

IT-01 Caught In/Between
IT-02 Caught Under
IT-03 Contact w/Temp. Extremes
IT-04 Exposure to Caustics
IT-05 Exposure to Noxious Fumes
IT-06 Exposure to Toxic Material
IT-07 Falls/Slips/Trips- Diff. Level
IT-08 Falls/Slips/Trips-Same Level
IT-09 Holding/Carrying
IT-10 Lifting-Load
IT-11 Lifting-No Load
IT-12 Overexertion
IT-13 Pushing/Pulling
IT-14 Reaching/Twisting
IT-15 Struck Against
IT-16 Struck By

BODY PART *(Back, Arm, etc.)*

BP-01 Abdomen
BP-02 Ankle
BP-03 Arms
BP-04 Back
BP-05 Chest
BP-06 Ear
BP-07 Elbow
BP-08 Eyes
BP-09 Face
BP-10 Finger
BP-11 Foot
BP-12 Genitals
BP-13 Hand
BP-14 Head
BP-15 Heart
BP-16 Hips
BP-17 Knee
BP-18 Leg
BP-19 Lungs
BP-10 Mouth
BP-21 Multiple Parts
BP-22 Neck
BP-23 Nose
BP-24 Shoulder
BP-25 Wrist

SOURCE OF INJURY *(Object or Substance that Caused the Injury or Illness)*

S-01 Animal/Insect
S-02 Biological
S-03 Chemicals
S-04 Clothing/shoes
S-05 Door/window
S-06 Dust
S-07 Electricity
S-08 Elevator/Escalator
S-09 Fire/Flame
S-10 Flying Object
S-11 Foliage/Tree
S-12 Food Products
S-13 Furniture
S-14 Glass
S-15 Hand Tool
S-16 Hot Objects
S-17 Ladder
S-18 Machine

S-19 Mechanical
S-20 Noise
S-21 Paint
S-22 Particles
S-23 Person
S-24 Power Hand Tool
S-25 Radiation
S-26 Sidewalk
S-27 Smoke/Fume/Vapor
S-28 Soaps/Detergents
S-29 Other
S-30 Steam/Hot Liquid
S-31 Utilities
S-32 Vehicle
S-33 Vibration
S-34 Walking Surface
S-35 Weather
S-36 Welding
S-37 Working Surface

UNSAFE CONDITION *(What Hazardous Condition Permitted the Occurrence of the Incident)*

USC-01 Congest Work Area
USC-02 Defective Tool/Equip.
USC-03 Excessive Noise
USC-04 Fire/Explosion Hazard
USC-05 Floors/Ramps
USC-06 Hazardous Atmosphere
USC-07 Hazardous Substance
USC-08 Improper Material Storage
USC-09 Inadequate Guard
USC-10 Inadequate Protection
USC-11 Inadequate Lighting
USC-12 Inadequate Ventilation
USC-13 Inadequate Warning System
USC-14 Machine
USC-15 Poor Housekeeping
USC-16 Radiation Exposure
USC-17 Stairs/Platform
USC-18 Temperature Exposure

A-17 Physical Limitation or Mental Attitude
A-18 Servicing Equipment in Motion
A-19 Unaware of Hazard
A-20 Unnecessary Task

CAUSE *(Management Control Function Which Led to the Incident)*

C-01 Improper Layout/Design
C-02 Inadequate Enforcement of Work Standards
C-03 Inadequate Environmental Control Program
C-04 Inadequate Hiring Standards
C-05 Inadequate Job Instructions
C-06 Inadequate Job Placement Standards
C-07 Inadequate Job Planning and/or Methods
C-08 Inadequate Maintenance Standards
C-09 Inadequate Preventive Maintenance Program
C-10 Inadequate Purchasing Standards
C-11 Inadequate Supervision
C-12 Insufficient Hazard Warning
C-13 Lack of Proper Job Procedures
C-14 Unsafe Design/Construction

UNSAFE ACT *(Substance, Behavior, or practice Which Permitted the Occurrence of the Incident)*

A-01 Did Not Use Proper Equipment
A-02 Failure To Get Help
A-03 Failure To Shut Down Power and/or Equipment
A-04 Failure To Use Guards Provided
A-05 Failure To Use Personal Protective Equipment
A-06 Horseplay
A-07 Improper Lifting, Lowering, Caring
A-08 Improper Load Placement
A-09 Improper Position For Task
A-10 Improper Use of Equipment
A-11 Influence of Alcohol/Drugs
A-12 Lack of Skill/Knowledge
A-13 Making Safety Device Inoperable
A-14 No Human Error
A-15 Operating At An Improper Speed
A-16 Operating Without Authority

A-21 Unsafe Act of Another Employee
A-22 Using Defective Equipment

SEVERITY OF INJURY

SVI-01 Disabling Injury (Perm)
SVI-02 Disabling Injury (Temp)
SVI-03 Fatality
SVI-04 First Aid Treatment Only
SVI-05 Medical Treatment
SVI-06 No Aid Required (Bruises)
SVI-07 No Injury Involved
SVI-08 Restricted Work