# Emergency Contact Numbers

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<tr>
<th>DESIGNATED OFFICE</th>
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<tr>
<td>Emergency Assistance (24/7)</td>
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<td>Department of Agriculture</td>
<td>734-3949</td>
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<td>Guam Fire Department (Barrigada, 24/7)</td>
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<td>Guam Memorial Hospital Authority</td>
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<td>Guam Regional Medical City</td>
<td>645-5500</td>
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<td>Guam Police Department (24/7)</td>
<td>472-8911</td>
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<tr>
<td>Poison Control Information Center</td>
<td>1 (800) 222-1222</td>
</tr>
<tr>
<td>University of Guam Plant Maintenance Office</td>
<td>735-2376 ext. 85</td>
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<td>University of Guam Student Health Services</td>
<td>735-2225</td>
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<tr>
<td>University of Guam Safety and Security Office</td>
<td>735-2370</td>
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<tr>
<td>University of Guam Safety Administrator</td>
<td>482-8671</td>
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<tr>
<td>University of Guam Security On-Duty</td>
<td>888-2456</td>
</tr>
<tr>
<td>U.S. Naval Hospital</td>
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Biosafety Manual

1 INTRODUCTION

University of Guam has developed this Biosafety Manual to implement the basic concepts of biological safety and promote the safe operation, handling and containment of infectious microorganisms and hazardous biological agents and materials used within the University’s laboratories and facilities and for any research operations conducted by our students, faculty and staff.

This Manual is applicable to all laboratory, research, service and support activities that may involve exposure to biological hazardous agents or materials including, and/or not limited to:

- work with recombinant and synthetic nucleic acids;
- microorganisms, such as bacterial, fungal, and parasitic agents;
- live viruses;
- human blood, body fluids, and tissues; and
- research animals.

The University’s safety program is based on the premise that every member of the community shares the responsibility for safety. As part of the community, it is important for laboratory personnel to be familiar with the health and safety guidelines that apply to their work and to conduct that work in the safest possible manner. All laboratory personnel must know and follow the procedures outlined in this manual. All operations performed in the laboratory must be planned and executed in accordance with the enclosed procedures. In addition, each employee is expected to develop safe personal chemical hygiene habits aimed at the reduction of chemical exposures to themselves, colleagues and students.

The information presented in this manual is not intended to be all inclusive. Departments, divisions or other work units engaged in work with potentially hazardous biological materials that have unusual characteristics, or are otherwise not sufficiently covered in this manual, must customize this document by adding additional sections addressing the hazards and how to mitigate their risks, as appropriate.

The Manual is mandated by federal and local regulations and guidelines. This Manual will be reviewed, evaluated and updated at least annually and is readily available to all employees, students, and their representatives.

2 PROGRAM ADMINISTRATION RESPONSIBILITIES

Responsibility for health and safety rests at all levels including the:
President of the University, who has ultimate responsibility for biological safety throughout the University and with the assistance of other program administrators will provide continuing support for chemical safety.

College Deans and Department Chairs have the responsibility for implementation of safe practices and procedures in their respective colleges or departments.

Principal Investigatory (PI): The PI has responsibility for implementation of safe practices and procedures in their laboratory, research grant or respective projects. The PI shall:

1. ensure that workers are trained and are in compliance with the Manual;
2. ensure that protective and emergency equipment is available, in working order, and that appropriate training has been provided;
3. ensure periodic laboratory inspections are performed;
4. know current legal requirements concerning regulated substances;
5. review and evaluate effectiveness of the Manual at least annually;
6. help project directors develop precautions and adequate facilities;
7. be able to identify potentially infectious and biohazardous materials and the risks associated with the proposed project; and
8. determine the proper level of personal protective equipment and its availability.

Biosafety Officer (BSO) shall:

1. assist PI’s and other laboratory employees with development and implantation of appropriate biosafety procedures and practices, including providing consultation and information;
2. keep abreast of regulatory requirements concerning regulated materials and communicate any changes to PI’s and laboratory employees;
3. review and evaluate effectiveness of the Manual at least annually and seek ways to improve the overall Manual;
4. help project, direct and develop precautions and adequate facilities;
5. monitor waste disposal program;
6. ensure that appropriate training has been provided to employees;
7. ensure that workers know and follow the Manual rules; and
8. maintain overall responsibility for the laboratory operations.

Institutional Safety Committee (ISC) shall work closely with the PI and is tasked with the responsibility to oversee and guide compliance with the Manual.

Instructor-in-Charge and Laboratory Supervisor shall:

1. monitor compliance with all safety regulations and practices within the individual laboratories or shops;
2. monitor that all students and employees receive appropriate training in the handling of hazardous chemicals and use of safety equipment;
3. respond appropriately to reports of unsafe conditions in the laboratory or shop; and
4. ensure that all necessary safety equipment is available and in working order.

Laboratory employees, faculty and students are responsible for:

1. planning and conducting each operation in accordance with practices and procedures established in the Manual;
2. using equipment only for its designed purpose;
3. being familiar with emergency procedures including knowledge of the location and use of emergency equipment for the laboratory, as well as how to obtain additional help in an emergency;
4. knowing the types of protective equipment available and using the proper type for each procedure; and
5. being alert to unsafe conditions and actions and calling attention to them so corrections can be made as soon as possible.

University of Guam units that are required to comply with the Manual are as follows:

- Cancer Research Center
- Center for Excellence in Development Disabilities Education, Research and Service (CEDDERS)
- Center for Island Sustainability
- College of Natural and Applied Sciences (CNAS) Division of Natural Sciences
- College of Natural and Applied Sciences (CNAS) Division of Agriculture and Life Sciences
- Cooperative Extension and Outreach
- Guam Aquaculture Development and Training Center (GADTC)
- Guam Ecosystems Collaboratorium (EPSCOR)
- Guam/Micronesian Area Health Education Center (G/M AHEC)
- Marine Laboratory
- School of Nursing and Health Sciences
- Sea Grant Program
- Water and Environmental Research Institute (WERI)
- Western Pacific Tropical Research Center (WPTRC)

Any department at University of Guam not listed above will also be required to implement the Manual if laboratory work with biological agents and materials are conducted.

3 DEFINITIONS

BBP: Bloodborne pathogens.

Biohazard: Biologically derived infectious agents that pose as a potential risk to human health or the environment.

BSC: Biological safety cabinet.

BSL-1: Biosafety Level 1 (Refer to page 11).

BSL-2: Biosafety Level 2 (Refer to page 12).

BSL-3: Biosafety Level 3.

BSL-4: Biosafety Level 4.

CDC: Centers for Disease Control and Prevention.

Containment: The action or policy of preventing the expansion of a harmful or infectious material and keeping it under control or within approved limits.

Decontamination: Any procedure that eliminates or reduces microbial contamination to a safe or acceptable level with respect to the transmission of infection.

Disinfectant: A chemical or a mixture of chemicals used to kill microorganisms, but not necessarily spores.

DOT: Department of Transportation.

FDA: Food and Drug Administration.

HEPA: High Efficiency Particulate Air.

IATA: International Air Transport Association.

Laboratory: A facility where the laboratory use of hazardous chemicals occur. It is a “workplace where relatively small quantities of hazardous chemicals are used on a non-production basis”.


OPIM: Other potentially infectious materials.

OSHA: Occupational Safety and Health Administration.
PPE: Personal Protective Equipment.

Sterilization: A process that kills and/or removes all classes of microorganisms and spores.

USDA: United States Department of Agriculture.

Waiver of Liability: A legal document that a person who participates in an activity may sign to acknowledge the risks involved in his or her participation. By doing so, the institution attempts to remove legal liability from the institution or person responsible for the activity.

4 BIOHAZARDOUS AND INFECTIOUS MATERIALS

4.1 CATEGORIES OF BIOHAZARDOUS AND INFECTIOUS MATERIALS

4.1.1 RECOMBINANT OR SYNTHETIC NUCLEIC ACIDS

The NIH defines recombinant and synthetic nucleic acids as:

1. Molecules that are constructed outside living cells by joining natural or synthetic DNA to DNA molecules that can replicate in a living cell (i.e., recombinant nucleic acids);
2. Nucleic acid molecules that are chemically or by other means synthesized or amplified, including those that are chemically or otherwise modified but can base pair with naturally occurring nucleic acid molecules (i.e., synthetic nucleic acids), or
3. Molecules that result from the replication of recombinant or synthetic nucleic acids.

Some examples of r-s-NA include plasmids, viral vectors, primers, and modified analogs of nucleotides.

The guidelines established by NIH specify containment levels and containment principles that are applicable to any recombinant DNA, RNA or other synthetic nucleic acid research or projects conducted in the laboratory. University of Guam complies with all aspects of biosafety practices presented by the NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules. The NIH exempts certain experiments that do not pose a significant risk to individual health or the environment. A full description of the exemptions with exceptions can be found in Appendix C of the NIH Guidelines. Refer to Appendix A on this Manual for the NIH Guidelines handbook.
4.1.2 HUMAN BLOOD, BODY FLUIDS, TISSUES AND BLOODBORNE PATHOGENS

OSHA Standard 1910 Subpart 1910.1030 (Bloodborne Pathogens Standard) minimizes or eliminates exposures to infectious agents that may be present in human blood, body fluids and tissues. The Bloodborne Pathogen (BBP) Standard applies to all employers who have employees that are “occupationally exposed” to human blood or other potentially infectious materials (OPIM) in the laboratory. An employee is considered to be occupationally exposed if there is reasonably anticipated skin, eye, mucous membrane, or parenteral contact with human blood or other potentially infectious materials that may result from the performance of the employee’s duties. An individual is also considered occupationally exposed if the employee uses equipment that is used to process or store blood, BBP or OPIM. All occupationally exposed employees are required by OSHA to attend a BBP training prior to beginning work and to maintain a valid certification of the training annually.

Bloodborne pathogens are pathogenic microorganisms that are present in human blood and can cause disease in humans. Examples of bloodborne pathogens include, but are not limited to, hepatitis B virus (HBV), hepatitis C virus (HCV), and human immunodeficiency virus (HIV). Other potentially infectious materials include human cell or tissue cultures, organ cultures, and bodily fluids, as well as blood, organs or other tissues from experimental animals infected with a bloodborne pathogen.

Refer to Appendix B for OSHA 29 CFR 1910.1030.

4.1.3 RESEARCH ANIMALS

Animal use and care is strictly regulated by several federal regulations. The Animal Welfare Act of 1969 (amended 1970 and 1976; P.L. 89-544, 91-579 and 94-279) (refer to Appendix D) provides broad protection to animals under the authority of the U.S. Department of Agriculture (USDA). It also regulates the transport, purchase and sale, care, handling, and use of animals in research and other purposes.

The Endangered Species Act of 1973 (P.L. 93-205) (refer to Appendix E) designates the Department of Interior (via the Fish and Wildlife Service) to protect both endangered and rare species, as well as habitats needed to sustain these organisms.

Caution shall always be taken when working with any animal. Users of laboratory animals must recognize that virtually all laboratory animal species can carry pathogens which are infectious to humans. Concern for the health of others who do not work directly with animals shall be paramount when laboratory animals are transported or used in general laboratory areas outside of an animal facility.

The basic considerations when working with animals include:

- Selection of an appropriate animal model
• Avoidance or minimization of pain or distress
• Surgical procedures and euthanasia
• Drug(s) which alter the animals awareness
• Chemical and physical restraints
• Alteration of life support systems
• Containment strategy for animal bio safety

4.1.4 TISSUE CULTURE AND CELL LINES

It is prudent to consider all cell lines potentially infectious. The most important element of safety in a cell culture laboratory is the strict adherence to standard microbiological practices and techniques. CDC and OSHA recommend that all cell lines or tissue cultures of human origin be handled at BSL-2. BSL-1 practices may be used for cell lines that are of non-human or non-primate origin and does not contain human or other primate pathogenic viruses and bacteria, mycoplasma or fungi.

5 PRINCIPLES OF BIOSAFETY

Biosafety is the application of knowledge, techniques and equipment to prevent personal, laboratory and environmental exposure or injuries from working with biological materials. The most important element in maintaining a safe work environment is strict adherence to good microbiological and laboratory practices and techniques.

5.1 BIOSAFETY LEVELS

There are four biosafety levels, with Biosafety Level 1 (BSL-1) being the least stringent and Biosafety Level 4 (BSL-4) being the most stringent. The Institutional Safety Committee (ISC) determines the proper biosafety level for working with a particular project. Currently, research at University of Guam is conducted at BSL-1 and/or BSL-2. University of Guam does not have specialized engineered laboratories to accommodate research at BSL-3 and BSL-4.

Below is a summary of each biosafety level. Detailed criteria for each level are described in Appendix F.

5.1.1 BIOSAFETY LEVEL 1

Bio-safety Level (BSL-1) facilities:

1. handle viable microorganisms not known to cause disease in healthy adults. These agents are often used in undergraduate teaching laboratories and work with these agents may usually be performed on laboratory benches.
2. represent a basic level of containment that relies on standard microbiological practices with no special barriers.
3. require a sink for handwashing.
4. have posted biohazard warning signs indicating BSL-1 on entrances.

5.1.2 BIOSAFETY LEVEL 2

Biosafety Level 2 (BSL-2) facilities:

1. handle viable microorganisms associated with human diseases of varying severity at moderate-risk. These agents can be hazardous through various exposure routes, but not inhalation.
2. represents all BSL-1 practices, in addition to limiting access to the laboratory.
3. have posted biohazard warning signs indicating BSL-2 on entrances.
4. require an autoclave for decontamination of infectious waste.
5. require work to be done within a biological safety cabinet to protect users from aerosols or splashes that can cause infection.
6. require special practices such as decontamination of all infectious material prior to disposal and implementation of an accident/incident plan that details exposure assessment and methods to clean up spills.

5.1.3 BIOSAFETY LEVEL 3

Biosafety Level 3 (BSL-3) facilities:

1. handle viable microorganisms associated with high-risk human diseases that are potentially lethal, and are hazardous through various exposure routes, including inhalation and ingestion.
2. represents all BSL-1 and BSL-2 practices, in addition to limiting access to the laboratory to only those personnel required for the program and who have been trained in the potential hazards of the specific agent in use.
3. have posted biohazard warning signs indicating BSL-3 on entrances.
4. have special engineering and design features including:
   a. self-closing doors and locks in accordance to institutional policies;
   b. separated areas for the laboratory, a clothing change room (anteroom), and unrestricted traffic flow within the building;
   c. hands-free or automatically operated sinks for hand washing;
   d. slip resistant floors that are impervious to liquids and chemicals;
   e. sealed seams, floors, walls, ceiling surfaces and windows;
   f. ducted air ventilation system;
   g. a readily available eyewash station;
   h. chairs that are covered with non-porous material.
5. require that the design, operational parameters and procedures must be verified and documented prior to operation. Facilities must be re-verified and documented at least annually.
5.1.4 BIOSAFETY LEVEL 4

Biosafety Level 4 (BSL-4) facilities:

1. handle viable microorganisms associated with high-risk, dangerous and exotic, life-threatening human diseases that are potentially lethal, for which there are no vaccines or treatments.
2. represent all BSL-1, BSL-2 and BSL-3 practices, in addition to limiting access to the laboratory to only those personnel required for the program and who have been trained in the potential hazards of the specific agent in use.
3. have posted biohazard warning signs indicating BSL-4 on entrances.
4. contain two models:
   1. A Cabinet Laboratory – manipulation of agents must be performed in a Class III BSC; and
   2. A Suit Laboratory – personnel must wear a positive pressure supplied air protective suit.
5. must provide laboratory personnel and support staff the appropriate occupational medical services including medical surveillance and available immunizations for agents handled or potentially present in the laboratory.
6. have special engineering and design features represented in BSL-3 facilities including:
   a. drains in the laboratory floor that are connected directly to the liquid waste decontamination system;
   b. liquid effluents from cabinet room sinks, floor drains, autoclave chambers, and other sources within the cabinet room must be decontaminated by a proven method before being discharged to the sanitary sewer;
   c. windows must be break-resistant and sealed;
   d. a dedicated non-recirculating ventilation system;
   e. decontamination of BSC must be performed using a validated gaseous or vapor method;
   f. a double-door autoclave;
   g. an appropriate communication systems between the laboratory and the outside.
7. require that the design, operational parameters and procedures must be verified and documented prior to operation. Facilities must be re-verified and documented at least annually.

6 PRIOR APPROVAL OF LABORATORY RESEARCH AND ACTIVITIES

Any persons seeking approval to conduct laboratory research activities involving humans or human tissues must get prior approval from the UOG Committee on Human Research Subjects (CHRS/IRB); and persons seeking approval to conduct laboratory research activities involving animals must get prior approval from the UOG Institutional Animal Care and Use Committee (IACUC). For additional information, please refer to
7 PERSONNEL TRAINING

Laboratory specific training is required on the part of the individual laboratory supervisor or Principal Investigator who shall instruct employees on procedures unique to the laboratory. This may include completion of a range of biosafety training programs prior to the initiation of work at the University of Guam.

8 PERMIT SYSTEM

A permit system shall be used for laboratory activities which present specific, foreseeable hazards. These activities include off-hours work, sole occupancy of building, hazardous operations, and unattended operations.

8.1 OFF-HOURS WORK PROCEDURES

Laboratory personnel when working after hours in the lab shall inform the Biosafety Officer or the Principal Investigator of planned activities.

8.2 SOLE OCCUPANCY

At no time shall work be performed in the laboratory when the only person in the building is the laboratory personnel performing the work. Under unusual conditions, crosschecks, periodic security guard checks, closed circuit television, or other measures may be taken when permitted.

8.3 HAZARDOUS WORK

All hazardous operations are to be performed during a time when at least two personnel are present at the laboratory. At no time shall a laboratory personnel, while working alone in the laboratory, perform work which is considered hazardous. The determination of hazardous operations shall be made by the laboratory supervisor and subject to permit.

8.4 UNATTENDED OPERATIONS

When laboratory operations are performed which will be unattended by laboratory personnel (continuous operations, overnight reactions, etc.), the following procedures will be employed:
• The permit shall be utilized.
• The laboratory supervisor will review work procedures to ensure for the safe completion of the operation.
• An appropriate sign will be posted at all entrances to the laboratory.
• The overhead lights in the laboratory will be left on.
• Precautions shall be made for the interruption of utility service during the unattended operation (loss of water pressure, electricity, etc.).
• The person responsible for the operation will return to the laboratory at the conclusion of the operation to assist in the dismantling of the apparatus.

8.5 UNDERAGE PERSONNEL AND “NON-EMPLOYEE” VOLUNTEERS AND VISITORS

Any person under the age of 18 will generally not be allowed to work in a laboratory where hazardous processes take place, or biohazardous materials are stored or used, unless approved by the Principal Investigator. All underage personnel must sign a Waiver of Liability form specific to the laboratory in which the individual will be working in. Underage personnel, including primary and secondary grade school students or visitors, must be directly supervised by faculty, staff or graduate students at all times. No key or key access shall be granted to underage personnel. This policy also applies to “non-employee” volunteers and visitors of the University of Guam.

9 STANDARD LABORATORY SAFETY PROCEDURES

9.1 GOOD WORK PRACTICES

9.1.1 GENERAL GUIDELINES

Each laboratory employee with the training, education and resources provided by supervision, shall develop and implement work habits consistent with this biosafety manual to minimize personal, student and coworker exposure to the biohazards in the laboratory.

9.1.2 SUPERVISION

Departments that utilize and maintain teaching/research laboratories and workshops on campus or conduct related activities at off campus sites are responsible for implementing applicable safety programs to ensure that these work areas are in full compliance with regulatory requirements. Departments must ensure that proper supervision is provided during University of Guam affiliated activities. Qualified supervisors need to be University of Guam employees who have attended applicable safety training/awareness programs. Qualified supervisors can include faculty, staff, and graduate students.
“Non-employee” visitors and volunteers, including minors, are permitted in the laboratories when there is a legitimate business or education purpose. A responsible person must be appointed by the Principal Investigator to supervise all visitors or volunteers when they enter a laboratory to work, or for a visit. Refer to Section 8.5.

9.1.3 PERSONAL HYGIENE

All laboratory personnel must know and follow the rules and procedures established in this manual.

- All employees shall remain vigilant to unsafe practices and conditions in the laboratory and shall immediately report such practices and/or conditions to the laboratory supervisor. The supervisor must correct unsafe practices and or condition promptly.
- Long hair and loose-fitting clothing shall be confined close to the body to avoid being caught in moving machine/equipment parts.
- Avoid unnecessary exposure to all biohazards by any route.
- Thorough hand washing is essential after handling any biohazardous material and prior to exiting the laboratory.
- Encourage safe work practices in co-workers by setting the proper example. Horse play is strictly forbidden.
- Do not eat or drink in the laboratory. Do not store food for human consumption in laboratory refrigerators.
- Seek information and advice from knowledgeable persons, standards and codes about the hazards present in the laboratory. Plan operations, equipment and protective measures accordingly.
- Use engineering controls of personal protective equipment in accordance with Section 9.2.
- Inspect personal protective equipment prior to use, and wear appropriate protective equipment as procedures dictate and when necessary to avoid exposure.

9.1.4 HOUSEKEEPING

Each laboratory worker is directly responsible for the cleanliness of his or her work space and jointly responsible for common areas of the laboratory. Laboratory management shall insist on the maintenance of housekeeping standards. Good housekeeping reduces risks and protects the integrity of biological experiments.

The following procedures apply to the housekeeping standards of the laboratory:

- The work area shall be cleaned and disinfected at the beginning and at the end of each operation and each shift.
• Work benches shall be kept as clutter-free as possible except those materials and equipment necessary for the work currently being performed.
• All floors, aisles, exits, fire extinguishing equipment, eyewashes, showers, electrical disconnects and other emergency equipment shall remain unobstructed.
• All apparatus shall be thoroughly cleaned and returned to storage upon completion of usage.
• All labels shall face front.
• Broken glassware shall be kept in the appropriate containers.

9.1.5 LABORATORY SIGNAGE AND LABELING

Each laboratory must have a room sign that provides safety information to individuals entering and working in the laboratory space. Room signs must contain designations for all laboratory hazards in use within the laboratory. The laboratory must also post an emergency contact card.

All biohazardous materials must be affixed with warning labels. The labels must appear on all equipment and containers that store, transport or ship biohazardous materials (i.e., bottles, refrigerators, waste containers, etc.).

9.2 PROTECTIVE CLOTHING AND LABORATORY SAFETY EQUIPMENT

9.2.1 GENERAL CONSIDERATIONS

The basic element of any personal protective clothing and equipment is the need to protect against the hazards in the workplace. Personal protective clothing and equipment must be provided at no cost to employees and students.

9.2.2 PROTECTION OF SKIN AND BODY

OSHA Standard 1910 Subpart (1910.132) requires the use of Personal Protective Equipment (PPE) in all labs where users are exposed to blood, chemical hazards, or mechanical irritants. Skin and body protection involves wearing protective garment and equipment over all parts of the body. PPE shall be selected on a task basis, and checked to ensure it is in good condition prior to use.

9.2.2.1 PROTECTIVE CLOTHING AND EQUIPMENT

Personal protective clothing and equipment may include impermeable aprons, face shield, gloves and shoe covers.

Lab coats are highly recommended to be worn in the laboratory. Laboratory coats shall be changed immediately upon discovery of significant contamination.
Closed-toe shoes must be worn in the laboratory at all times. Sandals, perforated shoes and bare feet are prohibited. Safety shoes, per ANSI Z47, are required where personnel routinely lift heavy objects.

Appropriate chemical-resistant gloves shall be worn based on the materials being handled, the particular hazard involved, and their suitability for the operation being conducted. Gloves shall be washed prior to removal from the hands to prevent skin contamination upon removal. Used gloves shall be inspected and washed prior to reuse. Non-disposable gloves shall be replaced periodically, depending on frequency of use and their resistance to the substances handled. Damaged deteriorated gloves shall be immediately replaced.

Thermal-resistant gloves shall be worn for operations involving the handling of heated materials and exothermic reaction vessels. Thermal-resistant gloves shall be non-asbestos and shall be replaced when damaged or deteriorated.

9.2.2.2 PROTECTION OF THE EYES

Safety glasses meeting ANSI Z87.1 are required for all personnel and any visitors where chemicals are handled and a chemical splash hazard exists. Safety glasses, goggles and goggles with a face shield shall be worn in the laboratory based upon the physical state, the operation or the level of toxicity of the chemical used.

Contact lenses can increase the risk of eye injury if worn in the laboratory thus contact lenses are prohibited in the laboratory, except as approved by the Chemical Hygiene Officer and supervisor.

9.2.2.3 PROTECTION OF THE RESPIRATORY SYSTEM

Respirator usage shall comply with the OSHA Respiratory Protection Standard, 29 CFR 1910.134. Refer to Appendix C.

9.2.3 LABORATORY SAFETY EQUIPMENT

9.2.3.1 BIOLOGICAL SAFETY CABINETS

The biological safety cabinet (BSC) is the primary means of providing containment and minimizing the exposures to hazardous biological materials caused by splashes and aerosols. BSCs are designed to provide individuals with environmental and product protection when used appropriately.

BSCs shall be located away from doors, heavily traveled laboratory areas, and other possible airflow disruptions.
BSCs control airborne contaminants during work with infectious material through the use of laminar airflow and High Efficiency Particulate Air (HEPA) filtration to sterilize and remove particulates from the air.

Often BSCs are equipped with germicidal ultraviolet (UV) lamps to decontaminate the interior surface. To ensure its efficacy, the lamp’s output intensity must be tested periodically, and the intensity shall not be less than 40 microwatts per square centimeter (uW/cm²) at the center of the work area. It shall be noted that UV lamps installed inside BSCs have limitations and users shall not rely solely on UV lamps for decontamination of BSC surfaces. Additionally, exposure to UV light may cause eye damage, so the proper PPEs must be worn. The skin must be covered. Glasses shall wrap around the face and shall comply with ANSI Z87.1.

BSCs are categorized into three types with specific performance characteristics. These types are: Class I, Class II, and Class III. Detailed criteria for each level are described in Appendix F.

BSCs shall be checked if it is operational and has been certified within the last 12 months by the Biosafety Officer, but shall be inspected every 3 months by the laboratory supervisor. A record of each inspection shall be maintained by the BSO. If the date on the certification sticker is more than 12 months, contact the BSO immediately for action.

9.2.3.2 EYEWASHES AND SAFETY SHOWERS

Whenever the eyes or body may be exposed to injurious corrosive chemicals, quick drenching or flushing of the eyes and body must be available for emergency use. All laboratories in which bulk quantities of hazardous materials are handled shall have access to eyewash and safety showers. As with any safety equipment, these can only be useful if they are accessible, there:

- Keep all passageways to the eyewash and shower clear of any obstacle.
- The eyewash shall be checked routinely to be certain that water flows through it.
- Showers shall be checked routinely to assure that access is not restricted and that the pull chain/bar is within reach.
- The flow through the safety showers shall be tested periodically to ensure sufficient flow.

9.2.3.3 FIRE SAFETY EQUIPMENT

Fire safety equipment easily accessible in the laboratory must include a fire extinguisher and fire blankets.
9.3 RECOMMENDED WORK TECHNIQUES

9.3.1 PIPETTES AND PIPETTING AIDS

Pipettes are used to transfer a measurable amount of liquid from one container to another. Safe pipetting techniques are required to minimize the potential for exposure to hazardous materials. The following procedures shall be followed for pipetting:

- Mouth pipetting is strictly prohibited. A pipetting aid shall always be used.
- If working with biohazardous or toxic fluid, confine pipetting operations to a biosafety cabinet.
- Do not discharge materials from a pipette at a height. When transferring liquids from one container to another, delivery shall be done with the tip of the pipette resting against the container to allow the fluid to flow down the container’s surface.
- Biohazardous mixtures shall not be prepared by bubbling air through the liquid with the pipette.
- Do not forcibly expel biohazardous fluid from pipettes.
- Avoid accidental release of infectious droplets.
- A towel wetted with disinfectant or a soft absorbent pad shall be used to clean up spills. The towel or pad shall be autoclaved after use.
- Discard contaminated disposable pipettes in an appropriate container.

9.3.2 SYRINGES AND NEEDLES

Syringes and needles shall only be used for parenteral injection and aspiration of fluids from humans, laboratory animals and diaphragm bottles. Use extreme caution to avoid accidental injection and the generation of aerosols during the use and disposal of syringes and needles. The following precautions shall be followed for using syringes and needles:

- Never leave an uncapped needle with the tip exposed unattended.
- Needles shall not be bent, sheared, or removed from the syringe.
- Never attempt to recap a needle.
- All used syringes and needles shall be promptly discarded into an appropriate sharps waste container.

In the event of a needlestick injury, wash the area thoroughly with soap and water. Notify your Principal Investigator, supervisor or instructor immediately. Individuals may report to Student Health Services or their healthcare provider of choice.

9.3.3 CENTRIFUGE EQUIPMENT

To minimize the risk of mechanical failure, centrifuges must be maintained and used according to the manufacturer’s instructions. To minimize the generation of aerosols
when centrifuging or homogenizing biohazardous material, the following precautions shall be followed:

- The work surface must be level and firm.
- Use safety centrifuge cups or a designated enclosed container to prevent aerosols from being released during centrifugation or homogenization of infectious material.
- Tubes and containers shall always be securely capped.
- Examine containers for cracks or stress marks before use. Discard any cracked containers.
- Always balance the tubes in the rotor before centrifugation. The buckets shall be paired by mass. Distilled water or alcohol (propanol, 70%) shall be used for balancing empty buckets.
- Do not open the lid while the rotor is moving.
- Clean the centrifuge and the rotor after every use.
- If any spills or leakage occurs during centrifugation, the centrifuge and the rotor shall be cleaned with a mild detergent such as 10% bleach, rinsed thoroughly with distilled water, and allowed to air dry completely.

9.3.4 BLENDERS, SONICATORS AND SHAKERS

Blenders, sonicators, and mixers are used for cell disruption, nanoparticle dispersion, homogenization and sample preparation. The following precautions shall be followed when using blenders, sonicators, and shakers:

- Domestic blenders are not recommended to be used in laboratories.
- Pressure builds up in the vessel during the operation of blenders, sonicators, and shakers, so contents shall be given time to settle before opening.
- Containers shall be opened in a biological safety cabinet to prevent the release of aerosols.

9.3.5 BUNSEN BURNERS AND LOOP STERILIZERS

A Bunsen burner produces a hot flame by combining flammable gas with controlled amounts of air. Bunsen burners are commonly used for heating and sterilization. Other flame-free alternatives include micro-incinerators or infrared sterilizers. The following precautions shall be followed when using Bunsen burners and loop sterilizers:

- Keep all flammable chemicals away from the work area in which a Bunsen burner will be in use. All flammable chemicals must be tightly sealed in the presence of an open flame.
- When using ethanol for sterilization, the dipping container holding the ethanol must be a stable glass container. Keep a non-flammable removable lid or a watch glass nearby to cap the container of ethanol in between use. Keep all ethanol containers at least a 30 cm distance away from the Bunsen burner.
Do not wear synthetic fabrics which are flammable and can melt to your skin.
Disposable transfer loops shall be placed in disinfectant after use and discarded
as contaminated waste.

9.3.6 WATER BATHS

Water baths are ideal when you need to maintain constant temperature over a period of
time. It is used to warm reagents, melt substrates, or incubate samples. The following
precautions shall be followed when using a water bath:

• Fill water baths with distilled water only.
• Do not leave water baths on overnight or when they will be left unattended for an
  extended period of time.
• To prevent electrical shocks, unplug the unit before filling or emptying.
• Ensure regular cleaning and disinfection of water baths.
  o Add an appropriate disinfectant, such as phenolic detergent, to water.
  o Raise the temperature to 194°F (90°C) or higher for 30 minutes once a
    week for decontamination purposes.
  o Do not use sodium azide for preventing growth of organisms. Sodium
    azide forms explosive compounds with some metals.

9.3.7 REFRIGERATORS, DEEP FREEZERS AND DRY ICE CHEST FREEZERS

Preservation of biological samples rely on maintaining proper storage temperatures to
keep the specimen intact. Scientific refrigerators and ultralow-temperature freezers
maintain internal temperatures for long-term sample storage due to efficient insulation
and cabinet or door-mounted gaskets. The following precautions shall be used when
using refrigerators, deep freezers, and dry ice chest freezers:

• To minimize breakage and leaks, place primary containers such as test tubes
  inside secondary containers prior to storage in freezing units.
• Refrigerators and freezers shall be connected to backup power systems in the
  event of power failures. Liquid nitrogen or liquid carbon dioxide systems can be
  used as alternatives.
• Refrigerators, deep freezers, and dry ice chests shall be checked, cleaned out,
  and defrosted periodically to remove any ampules, tubes, etc., containing
  biohazardous materials that may have broken during storage.
• For electrical safety, shut down units before proceeding with decontamination.
• All containers stored in refrigerators and freezers shall be clearly labelled.
  Unlabeled and obsolete materials shall be autoclaved and discarded.
• All inventory must be maintained of the refrigerator and freezer’s contents.

9.4 DECONTAMINATION

UOG Biological Safety Manual
All procedures and policies are subject to change and amendment. Refer to the UOG Policy and Procedure
Library website (https://www.uog.edu/policy-procedures-library/) for the official, most recent version.
The purpose of decontamination is to make a hazardous material, device, instrument or surface safe for further handling. A decontamination procedure can vary from simple cleaning with disinfectant to sterilization methods. There are four main categories of physical and chemical decontamination – liquid disinfection, use of heat, vapors and gases, and ultraviolet (UV) radiation.

9.4.1 LIQUID DECONTAMINATION

Liquid decontamination is generally used for surface decontamination. There are several commercially available disinfectants. Disinfectants shall be carefully selected based on the activity in which the specific agents are being used in the laboratory. The recommended general-purpose disinfectant solution is household bleach (5.25% sodium hypochlorite) diluted in 1:10 in water. A fresh solution of 10% bleach must be prepared daily. Each solution container must be labeled with a make-up date.

Ethanol or isopropyl alcohol (70% to 85%) is not a very effective disinfectant in most conditions because they lack sporicidal activity, can’t penetrate through protein-rich materials, and does not have the ability to kill hydrophilic viruses.

The following criteria shall be considered when choosing a disinfectant:

- The disinfectant must be effective against all organisms present in the laboratory.
- The disinfectant must be able to maintain sufficient contact time to inactivate the organism.
- The disinfectant must have sufficient stability in its diluted form to achieve inactivation.

9.4.2 HEAT

There are two types of heat sterilization – wet and dry heat.

Wet heat is the most dependable method of sterilization. Autoclaving, also called steam sterilization, is the most convenient method available in laboratories for rapidly decontaminating biological waste and sterilizing glassware, tools and media. Autoclaves use saturated steam under pressure of approximately 15 pounds per square inch (psi) to achieve a temperature of 250°F (121°C) for a prescribed amount of time. The following precautions shall be followed for safe and effective use of an autoclave:

- Use heat resistant gloves and face protection when removing processed material.
- Allow for the pressure to drop to 0 psi and the temperature to fall below 176°F (80°C) before opening the door. Crack the door slowly and wait a few minutes before fully opening it.
- Do not tightly cap bottles and test tubes. Autoclave bags shall be closed loosely to allow steam to penetrate.
Sterilization shall be verified routinely using biological (Bacillus stearothermophilus spores) or chemical (autoclave tape) indicators placed throughout the autoclave. B. stearothermophilus spore vials will display a lack of turbidity to indicate that the autoclave is achieving sterilizing conditions. Autoclave tape changes from beige to black to indicate when a temperature of 250°F (121°C) has been reached.

The autoclave shall be drained and cleaned daily.

Boiling does not kill all microorganisms and pathogens, but it may be used as the minimum processing for disinfection where other methods of decontamination are not available.

Dry heat uses temperatures of 320°F (160°C) or higher for 2 to 4 hours to sterilize laboratory tools and equipment. It is considered to be less efficient than wet heat and requires longer periods and higher temperatures to achieve sterilization. Burning or incineration are methods of dry heat sterilization. Incineration is useful for disposing of animal carcasses as well as anatomical and other laboratory waste.

9.4.3 VAPORS AND GASES

When employed in closed systems and with controlled conditions of temperature and humidity, vapors and gases are very effective disinfectants. Vapors and gases are primarily used to decontaminate biological safety cabinets, but they can also be used for building and room decontaminations. Due to their hazardous nature, contact the Safety and Security Office before arranging for vapor or gas decontamination methods.

9.4.4 ULTRAVIOLET (UV) RADIATION

Ultraviolet (UV) radiation is sometimes used in biological safety cabinets for surface decontamination. At a wavelength of 250 to 270 nanometers, UV radiation is a practical method for inactivating airborne viruses, mycoplasma, bacteria, and fungi. However, the usefulness of UV radiation limited by its low penetrating power thus dusty or soiled areas restrict its effectiveness. The following precautions shall be followed when using UV radiation for decontamination:

- Activate UV lights only when the area is unoccupied.
- UV radiation can cause serious burns to the eyes and the skin of individuals exposed for a short period of time, so proper shielding must be maintained when UV lamps are in use.
- UV lamp intensity and its destructive power decreases with time thus proper maintenance is critical for decontamination purposes. The lamp’s output intensity shall be tested periodically with a UV meter. Clean UV lamps every few weeks to prevent dust and dirt from accumulating.
9.5 CLEAN-UP OF BIOHAZARDOUS SPILLS

The appropriate spill response depends on the nature of the spilled biohazardous agent, the size of the spill and the location of the spill. The following sections outline suitable approaches to handling spills inside and outside a biological safety cabinet. Always wear personal protective clothing when clearing up spills. In the event of exposure of personnel to a biohazardous material, refer to Section 10.

9.5.1 BIOHAZARDOUS SPILLS INSIDE A BIOLOGICAL SAFETY CABINET

Chemical decontamination procedures shall be initiated immediately while the biological safety cabinet continues to operate to prevent escape of contaminants from the cabinet. The following procedure shall be followed during a biohazardous spill inside a BSC:

1. Spray or wipe walls, work surfaces, and equipment with an appropriate disinfectant.
2. Saturate the top work surface with disinfectant – applying disinfectant concentrically from the outer margin of the spill area, working toward the center – and allow to stand for 20 minutes to allow for adequate contact time.
3. Wipe up and remove excess disinfectant with a sponge or paper towel. Discard soaked towels or sponges in a biohazard bag.
4. After clean up, allow the cabinet blower to run for 10 minutes.
5. Inspect your clothing and exposed skin for contamination.

9.5.2 BIOHAZARDOUS SPILLS OUTSIDE A BIOLOGICAL SAFETY CABINET

For minor spills (less than 1 liter in size):

1. Cover the spill with paper towels to contain it.
2. Pour an appropriate disinfectant over the paper towels and the surrounding area. Apply disinfectant concentrically beginning at the outer margin of the spill area, working toward the center. Let stand for 20 minutes to allow for adequate contact time.
3. Wipe up and clear away soaked paper towels and transfer contaminated materials into a biohazard bag.
4. Inspect your clothing and exposed skin for contamination.

For major spills (over 1 liter in size):

1. Evacuate the area if there is a risk of exposure to aerosols. Allow aerosols to settle for 30 minutes.
2. Notify the supervisor, and warn others not to enter the contaminated area. Post a temporary warning sign on the door.
3. Pour an appropriate disinfectant around the spill and allow it to flow into the spill. To minimize aerosolization, avoid pouring the disinfectant directly onto the spill.
Paper towels soaked with the disinfectant may be used to cover the area. Let stand for 20 minutes to allow for adequate contact time.

4. Wipe up and remove excess disinfectant with a sponge or paper towel. An autoclavable dust pan, squeegee, and forceps for sharp materials may be used to transfer contaminated materials into a biohazard bag.

5. Wash adjacent area and spill area with an appropriate detergent solution.

6. Inspect your clothing and exposed skin for contamination.

9.6 BIOHAZARDOUS WASTE MANAGEMENT

The primary responsibility for identifying and disposing of biohazardous materials rest with Principal Investigators or laboratory supervisors. This responsibility cannot be shifted to inexperienced or untrained personnel.

Biohazardous waste include:

- Cultures and stocks of infectious agents and associated biologicals, cultures from medical, pathological, research and industrial laboratories, wastes from the production of biologicals, discarded and attenuated vaccines, and culture dishes and devices used to transfer, inoculate, and mix cultures
- Pathological wastes including tissues, organs, body parts and fluids removed during surgery, autopsy or other medical procedures
- Blood and blood products and items saturated and/or dripping with blood
- Animal carcasses, body parts and bedding of animals that have been exposed to infectious agents
- Sharps

Biohazardous waste must be properly handled and disposed of in order to minimize the risk of transmitting infection or endangering human health. Decontamination of wastes and their disposal are closely interrelated. The overriding principles is that infectious materials shall be autoclaved or otherwise decontaminated prior to disposal. Untreated waste must be carefully packaged to eliminate the chances of exposure during transport to its ultimate treatment and destruction site.

Any sharps or materials, including but not limited to hypodermic needles and syringes, Pasteur pipettes, broken glassware, capillary tubes, scalpel blades, disposable razors, and suture needles, contaminated with infectious agents that may puncture a bag must be deposited in a sharps disposal container. Sharps containers must be non-breakable, leakproof, impervious to moisture, tightly lidded, puncture resistant, and marked with the universal biohazard symbol.

9.7 SHIPPING, RECEIVING AND TRANSPORTATION OF BIOLOGICAL MATERIALS
The Centers for Disease Control and Prevention (CDC), the United States Department of Agriculture (USDA), the Food and Drug Administration (FDA), the Department of Transportation (DOT), the United States Postal Service and the International Air Transport Association (IATA) regulate the importation, shipment, and transportation of biological agents. It is the responsibility of the sender, carrier and the recipient to ensure that all regulations are enforced, that all requirements have been met, and proper documentation is provided.

9.7.1 PERMITS FOR TRANSPORT AND USE

Many biohazardous materials require federal permits for the transport and use of those agents. Refer to Appendix G for all recommended readings listed in the following passages.

The Department of Health and Human Services, through the CDC, and USDA established a set of regulations that require facilities and institutions to be registered and approved in order to transfer or receive biological agents and toxins. These biological materials include pathogenic bacteria or viruses, toxins from biological sources, blood or tissues capable of containing pathogens transmissible to humans and certain animals, and insects that may harbor disease-causing organisms. The current list of restricted agents and toxins under this rule can be found in Appendix F. Personnel working with select agents and toxins must go through the Bioterrorism Security Risk Assessment conducted by the FBI. Refer to USDA Agricultural Bioterrorism Protection Act of 2002; Possession, Use and Transfer of Biological Agents and Toxins (7 CFR 331) for more detailed information.

The USDA, through its Animal Plant Health Inspection Service (APHIS), regulates the import and transport of materials that could potentially harm US agricultural products, such as livestock or crops. APHIS requires permits for the importation and transportation of animal and plant pathogens, soil samples, insects, animals or animal products, plant or plant products, and genetically modified organisms into the environment. Refer to USDA Import-Export Regulations (9 CFR 300-399) and USDA Introduction of Genetically Engineered Organisms (GMO) Regulations (7 CFR 340.0-340.9) for more detailed information.

All biohazardous material intended to be shipped and transported must in accordance with the DOT Hazardous Materials Regulations (49 CFR 100-185) and IATA Dangerous Goods Regulations. To comply with DOT regulations, all biohazardous materials must be properly classified, packaged, documented, and handled by trained personnel.

9.7.2 ON-CAMPUS TRANSPORTATION

Biological materials transported between laboratories or buildings on campus must be contained to prevent release of materials into the environment. Transported materials must be in closed leak-proof primary containers secured with a tight cap. Plastic containers shall be utilized whenever possible. Primary containers shall be sealed in a
secondary container with absorbent material to contain liquids and absorb any shock during transport. Samples must be clearly labeled. The outer container must have a biohazard label attached as well.

10 EMPLOYEE HEALTH

10.1 ACCIDENTS AND EXPOSURE INCIDENTS INVOLVING BIOLOGICAL MATERIALS

All accidents, injuries and potential exposures involving biological agents must be reported. A report is necessary to insure proper care and treatment is given to those involved. Reports are documented by the supervisor of the work area where the event occurred. Additional reports shall be made directly to the individual's supervisor, Department, and Safety Office. Accidents, injuries, and exposures must be reported on the Worker’s Compensation forms for those employed by the University (refer to Appendix H). For students and volunteers, incidents can be reported on a standard Incident Report handled by campus security.

Any employee who sustains an exposure incident shall secure any potentially infectious material, remove all personal protective equipment and treat the affected area immediately. For life threatening injuries, call 911 or proceed immediately to the Emergency Department of Guam Memorial Hospital.

10.2 MEDICAL RESTRICTIONS

Employees must consult with an occupational physician to discuss medical conditions that can affect his/her safety in the laboratory. Pre-existing disease, medications, compromised immunity, and pregnancy or breastfeeding that may increase exposure to infants to certain agents, are some of the conditions that may increase the risk of an individual for a laboratory acquired infection.

It is recognized that exposure to certain infectious agents may adversely affect a fetus during pregnancy if the mother is infected with the agent. Thus women who are pregnant or become pregnant are encouraged to inform their Principal Investigators or supervisors.

10.3 MEDICAL CONSULTATION AND EXAMINATIONS

University of Guam will provide all employees who work with biohazardous materials an opportunity to receive medical attention. The opportunity for medical attention will be made available to employees under the following circumstances:
1. Whenever an employee develops signs or symptoms associated with a biohazardous chemical to which the employee may have been exposed in the laboratory,

2. Medical surveillance programs will be established where there are exposure monitoring and medical surveillance requirements, and/or,

3. Whenever an event takes place in the laboratory such as a spill, leak, explosion or other occurrence resulting in the likelihood of a hazardous exposure, the employee will be provided an opportunity for medical consultation for the purpose of determining the need for medical examination.

These medical consultations and examinations shall be provided without cost to the employees, without loss of pay and at a reasonable time and place.

These medical consultations and examinations shall be administered by or under the direct supervision of a licensed physician. Employees seeking the opportunity of medical consultation shall see Human Resources Office.
APPENDICES

A. NIH GUIDELINES FOR RESEARCH INVOLVING RECOMBINANT OR SYNTHETIC NUCLEIC ACID MOLECULES

B. OSHA BLOODBORNE PATHOGENS STANDARD 29 CFR 1910.1030

C. OSHA RESPIRATORY PROTECTION STANDARD 29 CFR 1910.34

Link: https://www.osha.gov/dte/library/respirators/major_requirements.pdf
D. ANIMAL WELFARE ACT OF 1969


E. THE ENDANGERED SPECIES ACT OF 1973

Link: https://www.fws.gov/endangered/esa-library/pdf/ESAall.pdf
F. BIOSAFETY IN MICROBIOLOGICAL AND BIOMEDICAL LABORATORIES, 5TH EDITION

Link: https://www.cdc.gov/biosafety/publications/bmbl5/BMBL.pdf

Link to Section IV – Laboratory Biosafety Level Criteria: https://www.cdc.gov/biosafety/publications/bmbl5/BMBL5_sect_IV.pdf

G. ADDITIONAL RESOURCES FOR SHIPPING AND TRANSPORTATION OF BIOLOGICAL MATERIALS

Link to Select Agents and Toxins List: http://www.selectagents.gov/SelectAgentsandToxins.html

Link to USDA Import-Export Regulations (9 CFR 300-399): https://www.ecfr.gov/cgi-bin/text-idx?SID=6bf96b7c000e93c9c49e1bdde46962be&c=ecfr&tpl=/ecfrbrowse/Title09/9cfrv2_02.tpl#300


Link to International Air Transport Association (IATA) Dangerous Goods Regulations: https://www.iata.org/publications/dgr/Pages/index.aspx
H. FACILITY SAFETY EQUIPMENT INSPECTION RECORDS

Facilities and safety equipment shall be inspected on a routine basis, every month. Inspections shall be conducted by the Biosafety Officer. The inspection shall be conducted by a competent person who is familiar with the specific contents of the Biosafety Manual and if possible familiar with applicable OSHA regulations. Facilities may choose to develop site specific inspection checklists. All records of inspections shall be maintained in this manual. Explanations shall be provided in writing when a significant delay is expected in corrective actions.
### FACILITY SAFETY EQUIPMENT INSPECTION RECORDS

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**Purpose:** “The purpose of this schedule is to assume that all laboratory safety equipment is inspected on a routine basis by competent personnel. Records in the form of inspection tags, stickers or logs will be maintained for all equipment.”

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**Chemical Hygiene Officer:** Telephone:
I. WORKER’S COMPENSATION COMMISSION FORMS

Link to Worker’s Compensation Commission’s (WCC) “What to Do In Case of A Work Injury”:

Link to Employee’s Notification of Injury, Illness, etc. Form:

Link to Employer’s Notification of Injury, Illness, etc. Form:

Link to Physician’s Authorization to Treat Injuries Form:
J. REFERENCES


K. ACKNOWLEDGEMENTS

The University of Guam would like to acknowledge Northeastern University in Boston, Massachusetts for the usage of their Biosafety Manual to use as a foundation and a base template for UOG’s Biosafety Manual.