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PREFACE

This manual was developed by the Institutional Biosafety Committee (IBC) of University Tun Hussein Onn Malaysia to provide information to protect workers and the surrounding environment and to comply with applicable standards and regulations.

The planning and the implementation of biohazard controls to prevent laboratory-associated infections and to control the spread of contamination must be a part of every laboratory activity in which biohazardous agents are used. The handling of biohazardous agents requires various precautionary measures depending on the agent(s) involved. The purpose of this manual is to provide general guidelines for evaluation, containment, and control of biohazards, categorized as degrees of risk of infection.

The implementation of this manual and procedures is the responsibility of the Principal Investigator and the person in charge of each laboratory. It is essential that they seek additional advice and training when needed to conduct research in a safe manner for employees, students, and the surrounding community. To assist in this regard, the Occupational Safety, Health and Environment Office services are available at the university.

SECTION 1: INTRODUCTION

1.1 PURPOSE OF MANUAL

This manual is prepared to explain the vital biosafety components of working in a research laboratory at University Tun Hussein Onn Malaysia (UTHM). This is to ensure students and staff have adequate information on training, safe work practices, safety equipment and personal protective equipment to protect them and the surrounding community from possible hazards associated with biohazardous agents.

Additionally, this manual includes an introduction to biosafety in Section 1, risk assessment in Section 2, procedures required for biohazard control in Section 3, emergency preparedness and response in Section 4, and guidelines for containment of biohazards in Section 5. This is for Principle Investigators (PI) who are required to obtain approval through the Institutional Biosafety Committee (IBC), UTHM and the National Biosafety Board (NBB). Lastly, this manual includes the control of biohazard of animals in Section 6:

1.2 DEFINITION OF BIOHAZARDS

The term ”biohazard” refers to any biological substance that is hazardous to humans, animals, or the environment. Numerous biohazards are comprised of valuable biological materials (VBM) VBM is defined as biological materials that require administrative oversight, control, accountability, and specific protective and monitoring measures in laboratories to protect their economic and historical (archival) value, as well as the population, from their potential to cause harm (according to their owners, users, custodians, caretakers, or regulators) as stated by World Health Organisations (WHO), 2006.

The VBM includes:

a. Pathogenic agents (bacteria, fungi, protozoa, parasites, sewage sludge isolates, etc.).

b. Recombinant or Synthetically Derived Deoxyribonucleic Acid (DNA), including those that are chemically or otherwise modified nucleotide analogues (e.g., morpholinos) or both. The definitions of Synthetically Derived DNA Molecules are:
    i. Molecules that (a) are constructed by joining nucleic acid molecules and (b) are able to replicate in living cells (i.e., recombinant nucleic acids);
    ii. Nucleic acid molecules that are chemically or otherwise modified but can pair with naturally occurring nucleic acid molecules (i.e., synthetic nucleic acids);
    iii. Molecules produced from the replication of those described in (a) or (b) above.
c. Recombinant DNA molecules, organisms, vectors (e.g., plasmids, viral vectors), and viruses containing recombinant DNA molecules.
d. Human and non-human primate blood, tissue, body fluid, and cell culture (primary and established cell lines).
e. Plants, animals, or derived waste that contain or may contain pathogenic hazards.

Some biohazards are unintended side effects of biologists working with or studying toxins or viruses. One common type of biohazard is clinica waste — things like used syringes or other tools contaminated withbacteria or other microorganisms. As biohazards have the potential to infect anyone exposed to them, all biohazard waste should be handled and disposed of properly.

1.3 OCCUPATIONAL SAFETY AND HEALTH COMMITTEE POLICY

The Occupational Safety and Health Act 1994 (OSHA 1994) provides a legislative framework to promote, stimulate and encourage high standards of safety and health at work. Act 514 contains a clear statement that '" *the responsibility for occupational safety and health at workplace is shared between the employers and the workers since the employers are the ones who provide the working environment which gives rise to hazards at work and the workers are the ones who have to work with these hazards*"'.

It is the policy of UTHM that in carrying out premises and/or worksites activities, we shall provide staff and customers, as far as is reasonably practicable, a safe and healthy work environment to ensure their welfare.

To ensure that this policy is enforced, it is the responsibility of the management to: -

a. **ENSURE** that the premises/worksites practice a safe and healthy work system.
b. **PROVIDE** facilities and basic equipment for them to conduct their daily duties safely.
c. **MAKE SURE** that all disseminated information, instructions, training, and supervision ensure staff safety.
d. **COMPLY** with the laws – Occupational Safety and Health Act 1994 (ACT 514) and all regulations and orders made thereunder.

1.4 RULES, REGULATIONS AND GUIDELINES GOVERNING THE USE OF BIOHAZARDS

On July 11, 2007, the Parliament of Malaysia passed the Malaysian Biosafety Act 2007. The Act came into force on December 1, 2009. The Act represents the fulfilment of Malaysia's international obligations as a Party to the Cartagena Protocol on Biosafety, ratified on September 3, 2003. The Biosafety (Approval and Notification) Regulations 2010 were passed and came into force on November 1, 2010 to implement the act. Together, they represent a new national scheme for the regulations of LMOs and products of LMOs.

The Biosafety Act 2007 describes the framework of the Malaysian regulatory system for LMOs and its products in which it is equipped with corresponding regulations and guidelines. The Act follows the standard structure of laws in Malaysia. The Act sets out the main features and represents the manner in which the provisions are to be implemented.

The Biosafety (Approval and Notification) Regulations 2010 facilitates the implementation of the Act and contains additional information about the operation of certain provisions in the Biosafety Act. The Regulations are enacted as a result of the powers conferred on the Minister by The Biosafety Act 2007. The Regulations govern matters such as the establishment of IBC, how an application for approval of release or import activity must be made, the duration for the decision to be made, matters relating to the certificate of approval, notification procedures for the export, contained use and import for contained use of LMOs, and appeal procedures against the decision of the NBB.

The Guidelines for Institutional Biosafety Committees (2010) operates in conjunction with the Biosafety Act and Regulations. The guidelines are the "operating instructions" issued by the Department of Biosafety (DOB) of the Ministry of Natural Resources and Environment (NRE) to provide guidance on the establishment of IBC, its role and functions and the processes that must be followed when obtaining, using, transferring, storing or destroying LMO/GMO materials. The guidelines state the responsibilities of the Biological Safety Officers (BSO) and researchers, IBC membership, reviews done by IBC, actions required to report incidents and spills and other related information.

The DOB has also issued the Biosafety Guidelines for Contained Use Activity of LMOs (2011). This guideline will help any organisation intending to carry out contained use activities involving LMOs and related materials to determine the Biosafety Levels (BSL) and facility type required.

The Act, Regulations and Guidelines can be downloaded from the Malaysian Biosafety website at http://www.biosafety.nre.gov.my or from the UTHM OSHE Office website at https://oshe.uthm.edu.my/v3 .

1.5 ROLES AND RESPONSIBILITY FOR COMPLIANCE

Any organisation conducting modern biotechnology research and development shall establish an IBC to ensure that every valuable biological material (VBM) including LMO/GMO, conducted at or sponsored by the organisation, irrespective of the source of funding, shall comply with the Malaysian Biosafety Act 2007 and any other related regulations and Malaysian laws relating to import and export, human, plant and animal health, environment and biological diversity.

1.5.1 Biosafety Unit

As for UTHM, Biosafety Unit is the authorised organisation to conduct surveillance audits related to evaluation, risk assessment, and invigilation of biosafety in laboratories involved in biological research or experiments, in line with the requirements of the Biosafety Act 2007.
Biosafety Unit is responsible for:

a. managing the registration of the IBC with the NBB.
b. managing the dissemination of biosafety policies and research-related issues such as valuable biological materials, laboratory safety, and other guidelines for researchers.
c. supporting the applications from UTHM researchers involved with LMO / GMO that require approval from the NBB.

1.5.2 UTHM Institutional Biosafety Committee (UTHM IBC)

UTHM IBC has been formed to accomplish a responsible Biosafety Unit. UTHM IBC should have the following minimum composition:

a. Chairperson.
b. Biosafety Officer (BSO) – a designated officer who helps ensure compliance with the Biosafety Act 2007 and the Biosafety (Approval and Notification) Regulations 2010 relating to LMO/GMO research conducted in the organisation.
c. Committee member.

The scopes of work of UTHM IBC are not limited to the following:

a. Provide guidance to Principal Investigator (PI) on biosafety policies and issues in LMO/GMO research, including the safety of laboratory personnel and other members of the organisation.
b. Recommend approvals for LMO / GMO research projects found to comply with the Biosafety Act 2007 and the Biosafety (Approval and Notification) Regulations 2010 and periodically review these research projects.
c. Assess and monitor the facilities, procedures, practices, training, and expertise of LMO / GMO research personnel.

   Notify the PI on the results of the IBC's reviews, approvals, or rejections of their applications and notify all activities involving the use of LMO / GMO to the NBB.
d. Assess and set containment levels for LMO / GMO research and modify them if necessary.
e. Assess field experiments to ensure that the proposed risk assessment, risk management, and emergency response plan are sufficient.
f. Adopt and implement an emergency response plan covering accidental spills and personnel contamination resulting from LMO / GMO research.

d. Review and report to the Head of the organisation and the NBB of any significant problems with non-compliance of the Biosafety Act 2007 and the Biosafety (Approval and Notification) Regulations 2010 and any research-related accidents or illnesses.
g. Ensure the information provided in the relevant application form (Approval/Notification) is correct and complete.

1.5.3 Research Centre /Laboratory

General biosafety training is mandatory for all individuals conducting research with LMO / GMO materials. Such training shall be organised by the organisation itself with guidance from the NBB. Individual researchers must provide proof to the IBC that they have undergone training or have adequate experience (as recognised by the IBC) in Biosafety and Good Laboratory Practices. This includes knowledge in handling and managing incidents/accidents in the facility and information on when and how to report laboratory incidents. Successful completion of training is recommended to receive IBC approval, either with a new application or for a re-application.

1.5.4 Principal Investigator (PI)

In general, PI shall understand relevant procedures, competent in risk analysis, able to develop risk management and Emergency Response Plan (ERP), as well as being competent in documentation.

Depending on the Risk Group that the biological agent may pose, the PI should conduct a risk assessment process, perform risk management procedures, and develop an emergency response plan (ERP) that includes the following:

a. Identify possible risks and assess the risks.
b. SOP for transportation of biological agents / LMO / GMO.
c. SOP for biological agents / LMO / GMO disposal.
d. SOP for waste treatment.
e. Additional locks on laboratory doors, freezers, etc., where biological agents / LMO/ GMO are used or stored.
f. Chain-of-custody forms within laboratories to track materials.
g. Inventories of biological agents / LMO / GMO.

h. Logs of access to areas where biological agents / LMO / GMO are used.

i. Written security plans for the use of biological agents / LMO / GMO which include:

   i. Procedures for access to biological agents / LMO / GMO.

      - Purchasing of microorganisms must be approved by BSO through the application of Request Order (RO) provided by Bursary UTHM. Refer **APPENDIX G**.

   ii. Procedures for routine cleaning, maintenance, and repairs.

   iii. Procedures for restricting unauthorised persons.

   iv. Procedures for addressing loss of keys, passwords and any other secured information and materials.

   v. Procedures for prevention of loss or theft.

SECTION 2: BIOLOGICAL RISK ASSESSMENT

Risk assessment is a process used to identify the degree of risk to laboratory workers, other personnel, and the environment. The degree of risk considers the virulence, pathogenicity, biological stability, and communicability of the organisms and the route of transmission. A biological risk assessment takes into account both the hazardous characteristics of the biological agents and the laboratory procedure hazards.

If the biological agents are GM, the risk assessment must take into account how the modification potentially changes the agents' hazardous characteristics such as virulence, pathogenicity, or susceptibility to treatments. This information can be found in Section 5: LMO / GMO Experimentation.

The laboratory manager or PI is responsible for ensuring that adequate and timely risk assessments are performed and for working closely with the Safety Committee and BSO to ensure that appropriate equipment and facilities are available to support the work being considered. Once performed, risk assessments should be reviewed routinely and revised when necessary, taking into consideration the acquisition of new data having a bearing on the degree of risk and other relevant new information from the scientific literature.

2.1 TYPES OF BIOHAZARD

2.1.1 Microbiological Waste

Microbiological waste is the most common type of biohazard waste; it refers to any laboratory waste that contains or has been contaminated with concentrated infectious agents.

2.1.2 Human Body Fluids

All fluids of the human body are in a liquid or in a semi-liquid state, including any and all products of human blood and blood, are a form of biohazard waste. Items that have been contaminated with blood, saliva, secretions, cerebral spinal fluid, amniotic fluid, peritoneal fluid, pericardial fluid,

pleural fluid, and/or synovial fluid in any way, shape, or form, are biohazardous. These wastes represent most of the hazardous waste.

2.1.3 Animal Wastes

Any animal carcass, body part, or bedding material that may have been used by an infected animal is also a form of biohazard waste. That is, unless the bedding material is inoculated with pathogenic microorganisms that are NOT infectious to humans. However, it is better to be safe than sorry in these cases and treat the bedding material as such.

2.1.4 Pathological Wastes

Pathological waste is any or all parts of the human body, organs, and tissues, including materials that may have come from surgical procedures, biopsy materials, or unfixed human tissue. Any of these may contain infectious agents. Waste materials from biopsy procedures fall into this category. Another example is the anatomical parts removed by the personnel during autopsies or surgeries should double-bag pathological waste to prevent leaks. Personnel should then store them in a secondary container to avoid liquid waste.

2.1.5 Solid Biohazardous Wastes

Solid waste is any non-sharp material that has contact with human or animal specimens. Such material includes personal protective equipment (PPE), petri dishes, towels, linens, and pipettes. Sharp materials such as scalpels and needles must be separated from other items, including fragile items. Other than that, blood vials and other glass objects become sharp when broken.

2.1.6 Liquid Biohazardous Wastes

Liquid waste such as body fluids or blood may contain an infectious agent. If the liquid is less than 25 millilitres, it can be disposed of as solid waste. Any amount exceeding 25 millilitres requires a different disposal method. Personnel must collect any liquid biohazardous waste in leak-proof

containers. They must secure the containers so that they do not overturn and label the containers as biohazards. For extra security, personnel can place the liquid containers in secondary containers, such as trays or buckets. Personnel can dispose of most liquid waste by treating it with bleach, or they can autoclave it as a liquid biohazard. An exception is a liquid that contains body fluid and chemical waste.

2.1.7 Sharp Biohazardous Wastes

Sharp biohazardous waste is any medical device that could be infectious and is sharp enough to puncture the skin. If it can puncture the skin, it can also puncture a plastic bag. This includes items such as needles, microscope slides, scalpels, and broken glass vials. Any of these may contain biohazardous material. Specific containers are designated for collecting sharp waste. These containers are resistant to puncture, leak-proof, and safe to handle. Personnel should collect all sharp waste in these special containers (Sharp bin). They should label the sharp containers with the correct symbol to identify them.

Figure 2.1 : Biohazardous Symbol

2.1.8 Genetically Modified Organisms (GMOs)

Risk assessments for working with GMOs should consider the characteristics of the donor and recipient/host organisms. Examples of characteristics to consider include the following:

a. Hazards arising directly from the inserted gene (donor organism).

Assessment is *necessary in situations where the product of the inserted gene has known biologically or pharmacologically active properties that may give rise to harm, for example:

i. Toxins
ii. Cytokines
iii. Hormones
iv. Gene expression regulators
v. Virulence factors or enhancers
vi. Oncogenic gene sequences
vii. Antibiotic resistance
viii. Allergens.

*The consideration of such cases should include an estimation of the level of expression required to achieve biological or pharmacological activity.

b. Hazards associated with the recipient/host.

i. Susceptibility of the host
ii. Pathogenicity of the host strain, including virulence, infectivity, and toxin production
iii. Modification of the host range
iv. Recipient immune status
v. Consequences of exposure.

c. Hazards arising from the alteration of existing pathogenic traits

Many modifications do not involve genes whose products are inherently harmful, but adverse effects may arise as the result of the alteration of existing non-pathogenic or pathogenic traits. Modification of normal genes may alter pathogenicity. In an attempt to identify these potential hazards, the following can be considered:

i. Is there an increase in infectivity or pathogenicity?
ii. Could any disabling mutation within the recipient be overcome due to the insertion of the foreign gene?

iii. Does the foreign gene encode a pathogenicity determinant from another organism?
iv. If the foreign DNA does include a pathogenicity determinant, is it foreseeable that this gene could contribute to the pathogenicity of the GMO?
v. Is treatment available?
vi. Will the susceptibility of the sewage to antibiotics or other forms of therapy be affected as a consequence of the genetic modification?
vii. Is eradication of the GMO achievable?

2.2 SOURCE AND ROUTES OF EXPOSURE

There are two modes of exposure at work to biological agents:

a. They are intentionally worked with, example in a microbiological laboratory; or
b. Incidental exposure may occur as a result of the kind of work done, such as healthcare work, farming, refuse disposal or work with products of animal origin. In the second group, the exposure to biological agents is incidental to the purpose of the work.

Factors that determine whether a person will contract a disease and how severe the condition will be include:

a. The contamination dose.
b. The classification of microorganisms by risk group.
c. The resistance (or susceptibility) of the individual immunity level and health background.

Some organisms can live outside a host for hours or even days, while others require a host to survive. Some organisms are tiny and lightweight, remaining in the air for long periods. Others quickly settle out onto surfaces and are a contact concern. All of these issues affect potential exposure.

Biological hazards can enter the body by different routes. When determining appropriate protective measures, a clear understanding of how biological hazards enter the body is an important first step.

Routes by which biological hazards can enter the body:

a. **Inhalation**; i.e., breathing.

i. Inhalation is a common way for biohazards to get into the body. The effect on the body depends on the biohazards and the amount they are inhaled.
ii. Although a person's immune system and lungs have mechanisms to fight germs, many biohazards are very strong and can overcome the defences.

Unlike chemical inhalation, people often do not realize that they have inhaled a bacteria, virus, or mold because there is no taste or smell and no irritating effects. It is only when there are symptoms of infection that a person becomes aware of any previous exposure.

b. **Absorption**; i.e., direct contact through breaks in the skin, even chapped skin, or through mucous membranes/contact with eyes, nose, mouth.

A biohazard can enter the bloodstream through broken skin, such as cut, chapped skin, hangnail, or any other break in the skin. Cover broken skin with a bandage or glove to seal the wound and wear appropriate protection to keep the wound area safe from biohazardous penetration. Splashes of blood/body fluids to the eyes is another way biohazard can be absorbed.

c. **Ingestion**; i.e., swallowing,

Swallowing biohazards can sometimes occur without us realising it, often from not washing our hands. Poor hand washing is one of the most common ways to transmit biological hazards. Workers should always wash their hands before eating so that any hazardous material on the hands is not ingested. **NO** drinking or eating in laboratories or any areas where specimens and other materials that are toxic by ingestion are commonly used. **NO** food should ever be stored in refrigerators where hazardous biological materials are stored.

d. **Injection**; i.e., through a puncture.

When something sharp punctures the skin, a biohazard can enter the body through the skin. In a laboratory, if the glass contains a biohazard, picking the pieces up with your bare hands could enable the biological agent to enter your body through the cut or puncture.

The most common routes of entry for biological hazards are inhalation and absorption from direct contact.

2.3 RISK GROUP AND BIOSAFETY CONTAINMENT LEVELS

In many countries, including Malaysia, biological agents are categorised into Risk Groups (RG) based on their relative risk. Depending on the country or organisation, this classification system considers the following factors:

a. Pathogenicity of the organism.
b. Mode of transmission and host range.
c. Availability of effective preventive measures (e.g., vaccines).
d. Availability of effective treatment (e.g., antibiotics).
e. Other factors.

WHO has recommended an agent RG classification for laboratory use that describes four general risk groups based on these principal characteristics and the route of transmission of the natural disease. The four groups address the risk to both the laboratory worker and the community.

a. **RG1** – is not associated with disease in healthy adult humans or animals.
b. **RG2** – is associated with the disease which is rarely serious and for which preventative or therapeutics is often available.
c. **RG3** – is associated with serious or lethal human disease for which preventative or therapeutics may be available.
d. **RG4** – is associated with lethal human disease for which preventative or therapeutics are not readily available.

It is recommended that all VBM used at UTHM be from Risk Group 1 and Risk Group 2, according to the biosafety level facility available.

In contrast to Risk Groups, Biosafety Levels (BSL) prescribes procedures and levels of containment for a particular microorganism or VBM. Similar to Risk Groups, BSL is graded from 1 – 4. Detailed descriptions of containment practices and biosafety levels can be found in the Laboratory Biosafety Manual, World Health Organisation (2004) and Biosafety Guidelines: Risk Assessment of Genetically Modified Microorganisms (2012). **Table 2.1** shows the relation of Risk group (RG) to Biosafety Levels (BSL), Pathogenicity features, practices, and equipment.

Table 2.1: Summary of the relation of Risk group (RG) to Pathogenicity features, Biosafety Levels (BSL), Practices and Equipment

RISK GROUP (RG)	PATHOGENICITY FEATURES	BIOSAFETY LEVEL**	LABORATORY PRACTICE	SAFETY EQUIPMENT
RG1 Low individual and community risk.	A microorganism that is unlikely to cause human disease of veterinary importance.	Basic- Biosafety Level 1. **Examples** Basic teaching, basic research laboratory.	Good Microbiological Techniques (GMT).	None, open bench work.
RG2 Moderate individual risk, limited to community, livestock or environment risk.	A pathogen that can cause human/animal disease but is unlikely to be a serious hazard to laboratory workers, the community, livestock, or the environment. Laboratory exposure may cause serious	Basic – Biosafety Level 2. **Examples** Primary health services, diagnostic services, research laboratory.	GMT plus protective clothing, biohazard sign.	Biosafety cabinet Class II for potential aerosols.

	infection. Effective treatment, preventive and control measures are readily available and can be implemented to control disease transmission. Risk of spread to a community is limited.			
RG3 High individual risk, low community risk.	Organism which may be an exotic or indigenous agent with potential to transmit disease mainly via aerosols. Disease caused is severe and may result in death. It could present a risk if spread in the community . However effective treatment, preventive and control measures are available.	Containment – Biosafety Level 3. **Examples** Special diagnostic services, research laboratory.	Level 2 plus special clothing. Controlled access, directional air flow.	BSC Class III and other primary devices for all activities.

RG4 High individual and community risk.	Organism which may be an exotic agent or new agent usually able to cause a life-threatening human disease. The infectious disease is readily transmissible from one individual to another. Infectious disease may be transmitted via aerosol or via an unknown route. Effective treatment, preventive and control measures are not readily available.	Maximum containment – Biosafety Level 4. **Examples** Dangerous pathogen units.	Level 3 plus airlock entry, shower exit, special waste disposal.	Class III BSC, positive pressure suits in conjunction with Class II BSCs, double ended autoclave (through the wall), filtered air.

****However, no one should conclude that the absence of an agent summary statement for a human pathogen means that it is safe to handle the agent at BSL-1, or without a risk assessment to determine the appropriate level of containment. The types of microorganisms in each risk group must be checked against a database that is updated on a regular basis. For instance, https://www.canada.ca/en/public-health/services/laboratory-biosafety-biosecurity/pathogen-safety-data-sheets-risk-assessment/actinobacillus aggregatibacter.html**

Majority of laboratories at UTHM provide Biosafety Level 1 (BSL-1) and Biosafety Level 2 (BSL-2) facilities. BSL-2 containment and practice are ideal for working with infectious agents to humans or animals where exposure can lead to limited to moderate disease.

2.4 RISK MONITORING

UTHM's research and teaching laboratories that work with biohazards are required to have annual inspections by the IBC UTHM using a laboratory biosafety checklist (refer to **APPENDIX D)**. Laboratory personnel should use this checklist to identify safety and regulatory deficiencies and address them before their annual inspections.

SECTION 3: PROCEDURES FOR BIOHAZARD CONTROL

3.1 FACILITY REQUIREMENTS

3.1.1 Biosafety Level 1 Laboratory (BSL-1)

The facility requirements of BSL-1 is presented in detail as the basis for all biosafety laboratories. Each laboratory should adopt safety or operation manuals that identify known and potential hazards and determine practices and procedures to eliminate or minimise such risks.
The facility requirements for BSL-1 laboratory are:

a. It is not separated from the general traffic patterns in the building.
b. It is designed with open benches which can be cleaned easily.
c. Bench tops should be impervious to water and resistant to acids, alkali, organic solvents, and moderate heat.
d. Laboratory furniture should be sturdy. Open spaces between and under benches, cabinets, and equipment should be accessible for cleaning.
e. Ample space must be provided for the safe conduct of laboratory work and for cleaning and maintenance.
f. Entrances to the laboratory should be labelled with appropriate signage identifying the type of laboratory facility and listing the procedures applicable, including emergency and maintenance procedures. The contact information of the laboratory supervisor or other responsible persons should be listed.
g. Space and facilities should be provided for the safe handling and storage of solvents, radioactive materials, and compressed and liquefied gases.
h. Equipped with a sink for handwashing.
i. May be equipped with a fume hood.
j. Autoclave or other means of decontamination should be available in appropriate proximity to the laboratory. Autoclaves should be certified annually by the Department of Occupational Safety and Health (DOSH) .Facilities for storing outer garments and personal items should be provided outside the laboratory working areas.
k. Facilities for eating, drinking, andresting should be provided outside the laboratory working areas.

l. A dependable supply of good quality water is essential. There should be no cross-connections between sources of laboratory and drinkingwater supplies. An anti-backflow device should be fitted to protect the public water system.

m. There should be a reliable and adequate electricity supply and emergency lighting to permit a safe exit. A stand-by generator is desirable for supporting essential equipment, such as incubators, biological safety cabinets, freezers, etc., and the ventilation of animal cages.

n. There should be a reliable and adequate supply of gas. Good maintenance of the installation is mandatory.

o. Safety systems should cover fire, electrical emergencies, emergency showers and eyewash facilities.

p. First-aid areas or rooms suitably equipped and readily accessible should be available.

3.1.2 Biosafety Level 2 Laboratory (BSL-2)

In addition to the facilities requirements for BSL-1, the following additional facilities are essential for BSL-2: -

a. Walls, ceilings, and floors should be smooth, easy to clean, impermeable to liquids, and resistant to the chemicals and disinfectants typically used in the laboratory. Floors should be slip-resistant.

b. A hazard warning sign incorporating the universal biohazard symbol and the level of containment together with access restrictions should be displayed on the access door to the laboratory work area. The hazard warning sign may also identify the agent and the name and telephone number of the Laboratory Supervisor or other responsible persons.

c. Windows in the laboratory should be closed and sealed.

d. Illumination should be adequate for all activities. Undesirable reflections and glare should be avoided.

e. Storage space must be adequate to hold supplies for immediate use and thus prevent clutter on bench tops and in aisles. Additional long-term storage space which is conveniently located outside the laboratory working areas, should also be provided.

f. Doors should have vision panels, appropriate fire ratings, and preferably be self-closing.

g. The laboratory should be well ventilated. If required, an inward flow of air should be maintained by extracting room air using mechanical ventilation to ensure directional airflow.

h. Freezers, refrigerators, or other storage units used for GM-BSL2 microorganisms located outside the designated laboratory should be lebelled with the appropriate signage.

3.1.3 Biosafety Level 3 Laboratory (BSL-3) and Biosafety Level 4 Laboratory (BSL-4)

BSL- 3 and BSL-4 require containment of airborne bacteria, viruses, or toxins; therefore, the design and administration of the BSL- 3 and BSL-4 facilities follow strict Biosafety Act and regulations with oversight provided by the NBB. BSL- 3 and BSL-4 are not available for use at UTHM; therefore, any activities involving RG3 and above are not allowed to be conducted at UTHM. For more information, please contact the Biosafety officer at OSHE UTHM or see the Biosafety Guidelines: Risk Assessment of Genetically Modified Microorganisms (2012).

3.2 LABORATORY PRACTISES AND TECHNIQUES

Good Laboratory Practice (GLP) embodies a set of principles that provides a framework within which laboratory studies are planned, performed, monitored, recorded, reported, and archived. GLP helps assure regulatory authorities that the data submitted is a true reflection of the results obtained during the study and can therefore be relied upon when making risk/safety assessments.

The objectives of GLP are to make sure that the data submitted is a true reflection of the results obtained during the study, data is traceable, and to promote international acceptance of tests.

3.2.1 Hazard Awareness Training

Employees should receive biological/chemical Hazard Awareness training for better understanding of the biological agents/chemicals they deal with and how they might be affected by such agents/chemicals while working or in the event of an accident. Proper segregation, handling, packaging, labelling, storage, treatment, and disposal of biological agents/chemicals are regulated by various laws to ensure that these processes are carried out safely.

In all regulations, there will be conditions for people doing the work to be trained on the nature of the hazards posed by the biological agents/chemicals. The purpose is to educate and understand the hazards

associated with people working with biological agents/chemicals. Biological/Chemical Hazard training provides the vital knowledge to make better risk assessments of the work and enables researchers to make the right choices about safe handling procedures, select the correct protective equipment, and make an effective and safe response to a spillage.

Biological and Chemical Hazard Awareness training courses should impart knowledge of the following:

a. Recognising hazard information.
b. Reading chemical names.
c. Understanding SDS and how to use data for risk assessments.
d. Predicting the behaviour of biological agents /chemicals if released.
e. Understanding how biological agents /chemicals can affect you and what poses an immediate danger.
f. Assessing risks.
g. Understanding the different types of protective clothing you may have to consider and a reminder that white lab coats are not universal protectors and etc.

Training may be accomplished through the IBC, informally in the laboratory setting, or through the formal training. Records of training sessions should be kept for each employee, along with an outline of the training programme and refresher training arranged according to the requirements.

3.2.2 Prohibited Activities

Basic safety rules:

a. A laboratory is a place where experimental work is carried out, and your full attention is required. For both safety reasons and to avoid disrupting laboratory activities, you must turn off all mobile phones; thus, their use is prohibited.
b. Everyone in the laboratory, including students, staff, and visitors, shall wear safety glasses, goggles, or face shields when there is potential for eye hazards. Goggles are recommended where chemical splashes are possible. If contact lenses are to be worn, the eyes should be protected by goggles when potential eye hazards exist. **NOTE**: prescription eyeglasses do not provide adequate protection.

c. Eating, drinking, chewing gum, and applying cosmetics are prohibited in the laboratory. Do not store food or beverages in the same refrigerators or freezers with chemicals, biohazards, or radioactive materials.
d. Appropriate gloves are essential when working with hazardous substances. All glove materials are not equally effective in protection from hazardous substances; consult a chemical resistance chart, a glove manufacturer, or OSHE UTHM for appropriate selection.
e. Never conduct unauthorised experiments or engage in horseplay in the laboratory. Please immediately report any unsafe behaviour to the instructor.
f. Wear appropriate clothing. In particular, you must wear closed-toed shoes (i.e., NO sandals or slippers) in the laboratory. Long pants are highly recommended. If you have long hair, tie it back, and scarves/ hijabs/ niqabs must be tucked into laboratory coats. Avoid wearing dangling jewellery.
g. Wearing an iPod, or any other device that interferes with hearing is not allowed.
h. Never pipet anything by mouth.
i. The work area must be kept clean and uncluttered. All chemicals should be labelled and stored properly.
j. Never work alone in the laboratory.
k. The hazards of chemicals used should be known (e.g., corrosiveness, flammability, reactivity, stability, and toxicity).
l. The Safety Data Sheet (SDS) should be available for all chemicals used and stored in the laboratory.
m. Always pay attention to your surroundings and be aware of what others are doing. Always be courteous.
n. Remove contaminated gloves before touching common use devices (door knobs, faucets, equipment); discard gloves before leaving the laboratory.
o. Always wash hands and arms with soap and water before leaving the laboratory.
p. Smoking is strictly prohibited.

3.2.3 Personal Protective Equipment (PPE)

PPE means any device or appliance designed to be worn or held by an individual for protection against one or more health and safety hazards. When working with biological agents/chemicals, PPE is worn

by workers to reduce or eliminate the exposure. The Safety Data Sheet (SDS) for chemicals in the laboratory or the workplace will list the correct types of PPE to wear. Not all types of PPE will protect against all hazards, so it is important to always check the SDS first before using both the chemical and the PPE. Please ensure all PPEs used in workplaces have a DOSH SIRIM PPE approval. The requirement for PPE suppliers/manufacturers to comply with the testing and certification requirements by SIRIM was introduced by DOSH in 2016.

The types of PPE that are commonly used are shown in **Table 3.1.**

Table 3.1: PPE Used for Body Protection

PPE	Body Part	Event
Safety Glasses	Eyes	Chemical liquid splashes, dust
Hard Hat	Head	Falling material
Ear Protection	Hearing	Excessive noise
Gloves	Hands	Corrosives, toxic materials
Respirator	Lungs	Toxic gases, vapours, fumes, or dust
Clothing	Skin	Toxic or corrosive materials
Footwear	Feet	Corrosive, toxic materials

PPE descriptions are classified according to body parts:

a. Body protection

i. Glove

Gloves are used to protect hands and, in some cases, parts of the arms from coming into contact with hazards. In the laboratory, the main hazards involve chemicals, biological material, radioactive materials, sharp objects, and extreme temperatures (autoclave, liquid nitrogen). The selection of gloves is based on the following factors to determine the best choice for each task**:**

- Chemical and physical hazards

  When working with chemicals, such as for cell cultivation medium, the best source of information on chemical hazards is a chemical resistance chart or database. It is available from a variety of manufacturers and contains information about the suitability of gloves for

use with chemicals. This information will guide the selection of glove materials and their thickness. The information available may not include every glove or compatibility data for every chemical. Manufacturers provide different degrees of information and may have different definitions of permeability, permeation rate, and degradation rating. They also use different rating systems making comparisons between manufacturers difficult at times. Material safety data sheets can also provide guidance on the selection of glove material.

Glove selection must also consider non-hazardous materials. For example, Polyvinyl Alcohol (PVA) gloves work very well for protection against some organic solvents but the coating is water-soluble and will quickly degrade when in contact with an aqueous solution.

Common physical hazards in the laboratory involve sharp objects or extreme temperatures. Leather and metal mesh gloves offer good resistance to bites, glass punctures, and cuts. Looped terry cloth gloves are a good option for handling autoclaved items assuming they are dry and cool.

- Potential contact time and splash hazard

  The higher the potential contact time or the risk of splash (by nature of the task or volume) requires higher protection through a thicker or longer glove. Immersing fingers, hands, or forearms into a hazardous material would require greater protection than incidental contact with small volumes. Pouring a substance would likely require more protection than pipetting work due to the risk of splashing during pouring.

- Dexterity

  A balance must be achieved when selecting gloves when hazardous materials are involved with precise tasks. A thicker glove may provide better protection, but a loss of dexterity may increase the risk of a spill.

ii. Disposable Gloves (small quantity handling)

Disposable gloves are thin (< 8 mil) and are most commonly made of latex rubber, nitrile rubber, polyvinyl chloride, or Neoprene. Disposable latex gloves typically offer sufficient

protection when handling small quantities or diluted chemicals with a low chance of contact or splash. They are not designed for applications involving prolonged, direct exposure to chemicals but rather for incidental splash exposures. Nitrile gloves provide protection from various types of hazardous materials and are more resistant to tearing. Disposable gloves must not be reused.

If chemical contamination occurs while wearing disposable gloves, immediately remove and discard the gloves. If contamination results from incidental contact (small amounts of chemicals that will dry quickly), remove the gloves and dispose of them in regular garbage. If gloves are grossly contaminated (were immersed in, saturated with, or are still wet with chemicals), they should be collected as hazardous waste in labelled plastic bags. After removing contaminated gloves, wash your hands and put on new gloves.

*** Gloves Use Restrictions:**

Gloves should not be worn in elevators, restrooms, libraries, conference rooms, cafeterias (or other eating areas), stairs, offices, or non- laboratory floors. If a person is wearing gloves, remove them before answering the phone or touching equipment or doorknobs. This will prevent contamination of facilities and other personnel if the gloves are contaminated. Conversely, it will prevent contamination of the gloves if they are being used to protect the material the personnel are handling. Use a secondary container to transport materials. It will allow the person to remove gloves as well as protecting the work.

iii. Lab coat

A laboratory coat or equivalent protection is required when working with or when working in close proximity to hazardous chemicals, unsealed radioactive materials, and biological agents at BL-2 or greater. A flame-resistant lab coat is required when handling pyrophoric substances outside of a glove box. It is recommended that a flame-resistant lab coat be worn when working with all flammable chemicals. Laboratory supervisors shall carry out a hazard assessment to identify situations (task, experiment, or area) in which alternative apparel or more protective apparel must be worn.

In general, protective clothing, including lab coats, should not be used as a substitute for engineering controls such as a fume hood, a glove box, process enclosure, etc., or as a substitute for good work practices and personal hygiene. For significant chemical handling, it will be necessary to supplement lab coat used with additional protective clothing, for example, a rubber or vinyl apron for handling large quantities of corrosives or hydrofluoric acid, or it may be preferable to use chemical-resistant coveralls for full body protection. Conversely, the use of engineering controls such as fume hoods does not preclude the need for wearing the proper PPE, including lab coats.

When lab coats are in use, the following should be observed:

- Wear lab coats that fit properly. Lab coats are available in a variety of sizes. Some lab coat services also offer custom sizes (e.g., extra-long sleeves, tall, or woman's fit). Lab coats should be fastened neatly close to the collar to provide optimal protection.
- Lab coats should be worn fully buttoned or snapped with sleeves down.

Wear lab coats only when in the laboratory or work area. Take off the lab coats when leaving the laboratory /work area.

<u>Action to be taken if lab coat is contaminated with:</u>

- **Biological agents (Blood, body fluids, or BSL-1 and BSL-2 agents)**: place the coat in an autoclave bag and report to the laboratory staff. The staff should seal the autoclave bag and autoclave it using decontamination conditions (121 degrees C, 20 mins,1 psi)* **Radioactivity**: Place the lab coat in a plastic bag, seal, label*
- **Chemical**: If the chemical can be seen and the contamination is minor, rinse with water. If it is gross contamination or contamination with a select carcinogen, place the coat in a plastic bag, seal, *label

  * the contaminants will be collected for management of scheduled waste by a registered contractor appointed by the university.

b. Eye protection

As a minimum standard, safety glasses/goggles must be worn whenever handling hazardous materials. The eyes and face should be protected from potential splashes when conducting work with hazardous substances in a chemical fume hood. Chemical splash goggles underneath a face shield must be worn when a chemical fume hood is not used as there is a potential for a hazardous substance to splash.

Reminder/Precaution:

Contact Lenses: If you choose to wear contact lenses when handling chemicals, be sure to wear safety glasses. In the event of a chemical splash to the eyes, contacts can hold the chemical in the eyes, prolonging the exposure and increasing eye damage.

c. Respiratory Protection

Laboratory ventilation and chemical fume hoods usually control exposure to hazardous chemicals. Laboratory technician will test, train, and issue a cartridge respirator to an employee if there is a potential for overexposure. Do not purchase respiratory protective devices of any kind without the laboratory technician's approval. If you believe a hazardous airborne exposure condition may exist in your work area, contact the person in charge of the laboratory.

d. Foot protection

People who are working in the laboratory must wear sturdy-soled, well-fitting shoes that cover the entire foot. Sandals, slip-ons, perforated shoes, and open-toe shoes are not acceptable in the laboratory. Leather shoes are recommended.

3.2.4 Biohazard Warning Door Sign

The Biohazard Warning door sign is displayed in addition to the biohazard symbol on the permanently affixed caution sign next to laboratory entry doors. The Biohazard Warning Sign (**Figure 3.1**) must be permanently affixed to entry doors of the following laboratories/rooms:

a. BSL-1

b. BSL-2

Figure 3.1: Biohazard Warning Sign for Laboratory Entry Doors

It is important that all employees, students and visitors adhere to the policy of entering areas where these signs are displayed, such as laboratories, cold rooms, warm rooms, chemical storage areas, and autoclave rooms. **Table 3.2** shows a list of laboratory door sign requirements. As for UTHM laboratories, technicians or laboratory managers can make a request to the IBC committee at their faculty or refer directly to the Biosafety Officer at OSHE.

Table 3.2: Laboratory door sign requirements

Laboratory Door Sign	Requirement
Do all entrances to the laboratory space have appropriate caution signs that indicate hazards in the area?	It should be visible to all pedestrians before entering hazardous areas.
Is the contract information complete, legible, and up to date?	The Principle Investigator responsible for the area in question must have the name and information of the 24-hour emergency contact written on the laboratory door sign. It is strongly recommended that an alternative contact also be written on the laboratory door sign.

3.2.5 Handwashing

Keeping hands clean is one of the most important steps to prevent infection and avoid getting sick and spreading germs or bacteria to others. Therefore, good handwashing is **MANDATORY** in laboratory practice, especially:

a. After handling biohazardous agents, chemicals or animals.
b. After removing gloves.
c. Before leaving the laboratory.

3.2.6 Good Housekeeping

Good housekeeping practices in the laboratory can significantly minimise the risk of accidents and exposure to hazardous materials. The practices include the following:

a. Bench tops shall be organised with large equipment in the back and progressively smaller equipment toward the front, with sufficient space to perform work safely.
b. Laboratory components that are not being used shall be placed in the designated place. Avoid accumulating large amounts of unnecessary items on the work benches.
c. Clean and tidy up laboratory equipment and apparatus when the experiment is completed. Never leave them unattended on the work benches.
d. Remove unnecessary items on floors, under benches, or in corners.
e. Regularly check glassware for star cracks, chips, or cracks, and promptly discard or repair any unsafe glassware.
f. Keep drawers and cabinets closed when not in use.
g. Spilled chemicals, biohazard materials, and any powdered materials shall be cleaned immediately to eliminate the risk of hazards.
h. Properly secure all compressed gas cylinders.
i. Properly dispose of biohazard and chemical wastes. Old and unused samples should be disposed of promptly and properly in accordance with the specified guidelines.
j. Remove any clutter that interferes with access to emergency equipment such as eyewash stations and fire extinguishers.

3.2.7 Inventory Control

Good inventory procedures can significantly help the management of laboratory stock. Inventory of infectious microbe needs to be controlled and closely monitored to minimise the risk of biohazards. Old and unused hazardous microbes have to be disposed of properly in accordance with the specified guidelines.

All of these inventory and disposal activities shall be updated and recorded systematically by including the important details such as the person in charge and the location of the materials (i.e. in a freezer, safety cabinet, or shelf).

3.2.8 Minimisation of Aerosols

Experimental procedures in the laboratory shall be conducted carefully to minimise the creation of aerosols. Some mitigation approaches include the following:

a. **Opening tube**

   i. Manipulate infectious materials within a biological safety cabinet.
   ii. Upon opening, unscrew the cap slightly and wait a few seconds before removing it.

b. **Pipetting**

   i. Use ‘to deliver’ pipettes calibrated to retain the last drop.
   ii. Use pipettes with plugs.
   iii. Discharge pipettes close to the fluid level and let the contents run down the wall of the container.

c. **Breakage**

   i. Use plastic tubes, flasks, and bottles where possible.
   ii. Use screw-capped tubes and bottles rather than plugs or snap caps.

d. **Inoculating loop**

   i. Use a micro-incinerator or a disposable loop instead of a Bunsen burner.
   ii. Allow the inoculating loop to cool before any procedures.

e. **Homogenising**

i. Ensure the laboratory blender has a gasket lid and leak-proof bearings.

ii. Wait a few seconds before opening the lid after mixing.

f. **Centrifugation**

i. Centrifuge infectious material in closed containers, place in sealed safety cups or rotors.

ii. Open cups in a biological safety cabinet.

iii. Wait five minutes before opening the centrifuge after each run to allow any aerosols to settle.

g. **Poring infectious materials**

i. Perform your work over plastic-backed absorbent material.

ii. Wipe the rim of the tube with disinfectant-soaked absorbent paper to remove potential contamination on the outside of the tube.

3.2.9 Risk Communication

Biological hazards are hazardous because they are not always physically visible to humans. Biohazard warning labels should be affixed as close as possible to containers with string, adhesive, or other methods that prevent their loss or unintentional removal. It is very important to warn laboratory users of hazardous materials by placing biohazard labels and tags on storage containers, waste containers, or anything that contains biological materials. Warning labels must also be affixed to biohazardous waste, containers, refrigerators, and freezers containing Other Potentially Infectious Material (OPIM), and other containers used to store, transport, mail, or ship the OPIM. In addition, a clear procedure for handling biohazards shall be visible to users, ensuring maximum protection and safety.

3.3 LABORATORY EQUIPMENT

3.3.1 Biological Safety Cabinets (BSC)

The BSC is designed to reduce the potential escape of research material into the worker's environment and to remove contaminants from the research work zone. BSCs provide a clean work zone (product protection), aerosol protection for the operator (personnel protection), and environmental protection through the use of High-efficiency Particulate Air (HEPA) filters. HEPA filters are effective at trapping particulates and infectious agents. They do not capture volatile chemicals or gases.

Other equipments such as horizontal laminar flow units, vertical laminar flow units, and non-ventilated tissue culture boxes are not biosafety cabinets. They provide only limited product protection and do not provide any personnel protection. If laminar flow units are present in the laboratory, they must be labelled as "**NOT for Use with Pathogenic Organisms**."

a. Types of Biological Safety Cabinets

i. **Class II Biosafety Cabinets**

(Equipment to protect the worker, product, and environment)

- Class II, Type A1 BSCs are suitable for work with low to moderate risk biological agents requiring BSL-1, BSL-2, or BSL-3 containment in the absence of volatile toxic chemicals and volatile radionuclides. The build-up of chemical vapours in the cabinet (by recirculated air) and in the laboratory (from exhaust air) could create health and safety hazards.

- Class II, Type A2 BSCs are suitable for work with low to moderate risk of biological agents requiring BSL-1, BSL-2, or BSL-3 containment. Minute quantities of volatile toxic chemicals or volatile radionuclides can be used in a Type A2 cabinet only if the cabinet exhausts to the outside via a properly functioning canopy connection.

- Class II, Type B1 BSCs are suitable for work with biological agents requiring BSL-1, BSL-2, or BSL-3 containment. They may also be used with biological agents treated with toxic chemicals and trace amounts of radionuclides required as an adjunct to microbiological

studies if the work is done in the direct exhausted portion of the cabinet or if the chemicals or radionuclides will not interfere with work when recirculated in the downflow.

- Class II, Type B2 BSCs are suitable for work with biological agents requiring BSL-1, BSL-2, or BSL-3 containment. They may also be used with biological agents treated with toxic chemicals and radionuclides required as an adjunct to microbiological studies. This type of cabinet is sometimes referred to as a "Total Exhaust Cabinet."

- Class II, Type C1 BSCs are capable of being configured in the recirculating Type A mode for standard microbiological work or may be connected to the building exhaust to function in the Type B mode for handling biological material with hazardous chemical vapours or radionuclides.

b. BSC Certification

i. Equipment must be decontaminated prior to maintenance work, repair, testing, moving, changing filters, changing work programs, and after gross spills.
ii. The methods and requirements for testing BSCs vary depending upon the design of the cabinet and its intended use.
iii. All research materials must be removed from the BSC prior to testing and certification.
iv. Plan and schedule in advance as the BSC cannot be used until certification is complete.
v. The University's IBC requires all BSCs to be tested and certified prior to initial use, relocation, after HEPA filters are changed, and at least annually.
vi. The testing and certification process include:
    - A leak test to assure that the airflow plenums are gas tight in certain installations.
    - A HEPA filter leak test to assure that the filter, the filter frame, and the filter gaskets are in place and free from leaks. A properly-tested HEPA filter will provide a minimum efficiency of 99.99% of particles 0.3 microns in diameter and larger in diameter. Measurement of airflow is to ensure that velocity is uniform and unidirectional.
    - Measurement and balance of intake and exhaust air.
    - Users must receive training prior to the use of BSCs. The training is the responsibility of the PI.

c. Basic Guidelines for Working in the BSC

i. Never place anything over the intake or rear exhaust grill. Keep equipment at least four inches inside the cabinet window and perform all transfer operations of viable material as deeply into the BSC as possible.
ii. Do not overload BSC with equipment and other items. Only bring in the items needed for work.
iii. Plan in advance to have all required equipment inside the BSC. Good laboratory technique minimises arm movements through the air barrier until the procedure is complete.
iv. During manipulations inside the BSC, segregate contaminated and clean items. Keep clean items out of the work area and place discarded containers to the rear of the BSC.
v. Avoid entering and exiting the workroom. Foot traffic can cause disruptive drafts that allow microorganisms to escape through the air barrier of the BSC.
vi. Equipment should be kept as parallel as possible to the downflow of the airstream.
vii. To purge airborne contaminants from the work area, allow the BSC to run following the completion of work. The BSC can be turned off after 20 minutes, but it is recommended to be left on continuously.
viii. Decontaminate the BSC after use. Choose a disinfectant that does not corrode the stainless steel surface or follow disinfection with an ethanol or water wipe to remove corrosive chemicals.
ix. Do not use an open flame Bunsen burner inside a BSC. If required, a touch-a-matic burner or infrared loop sterilizer should be used. An open flame Bunsen burner disrupts the unidirectional air stream. The flame could damage the filter or set fire to the BSC when the BSC is turned off.
x. Do not use the BSC for storage when it is not in use.

3.3.2 Autoclave

a. One of the most effective physical decontamination controls is steam sterilization (autoclave) which generates moisture and high temperature pressurised steam within a sealed chamber.
b. Autoclaves can sterilize all items that are heat stable. In gravity autoclaves, a cycle of 250°F (121°C) at 15 to 18 pounds per square inch (psi) of pressure for one hour may be required for

decontamination. In the newer vacuum autoclaves, decontamination may require a cycle of 270°F (132°C) at 27 to 30 psi for 45 minutes.

c. A biological indicator should be used to verify proper autoclave operation.
d. Personal protection equipment (PPE) such as rubberised aprons, full-face shields and heat and liquid-resistant gloves must be worn when operating autoclaves.
e. Position items in the autoclave to allow steam penetration into all items to be decontaminated.
f. Materials in tightly sealed or stoppered containers may not be effectively decontaminated and may become dangerously pressurised, causing injury when removed from the autoclave.
g. Items containing chemicals such as phenol or chloroform should never be placed in an autoclave.

3.3.3 Water Baths and Incubators

a. These equipment are closely linked to the hazard concerning the growth of microorganisms. The users must ensure regular cleaning and disinfection.
b. After usage, decontaminate water baths and incubators with appropriate decontaminants.
c. Do not use sodium azide to prevent the growth of organisms as sodium azide forms explosive compounds with some metals.
d. Maintenance service on water baths and incubators that appear to be improperly used and/or contaminated may be denied. It is not the responsibility of maintenance personnel to clean up after the usage of laboratory personnel.

3.3.4 Refrigerators, Deep Freezers and Dry Ice Chests

a. Refrigerators, deep freezers and dry ice chests should be checked and cleaned periodically to remove any broken ampoules, tubes, etc., containing biohazards.
b. Containers must be stored in proper order and sequence and properly labelled to preclude the withdrawal of the wrong ampoules or tubes.
c. The use of gloves and respiratory protection during the cleaning of refrigerators, deep freeze or dry ice chests is recommended.
d. All stored materials should be properly labelled with the scientific name, the date stored, and the name of the individual storing the material.

e. Flammable solutions that require 4°C storage conditions must be stored in a refrigerator approved for flammable storage.
f. Notices of this flammable effect should be placed on the refrigerator doors.

3.4 LABORATORY PROCEDURES

3.4.1 Pipetting

a. Delivery with the tip of the pipette resting against the container allows the fluid to flow down the surface and minimises aerosols.
b. Allowing a droplet to fall from the tip of a pipette, intentionally or accidentally, results in aerosol production, the extent of which depends on the height of the fall and the surface upon which the droplet lands. The following procedures should be followed for pipetting:
    i. Mouth pipetting is strictly prohibited. Mechanical pipetting aids must be used.
    ii. Infectious mixtures should not be prepared by bubbling air through the liquid with the pipette.
    iii. Infectious materials should not be forcibly discharged from pipettes (e.g., the last drop forcefully removed).
    iv. A towel wetted with disinfectant or a soft absorbent pad covering the immediate work surface is most useful in absorbing droplets and small spills.

3.4.2 Using Needles and Other Sharp Items

a. Sharp items are used to cut or puncture skin or body parts, including needles, scalpels and lancets.
b. Other sharp items can still cause injuries, although they do not fit the regulatory definition of sharp, such as broken glass, glass septum vials, glass pipets, razor blades, and sharp teeth and nails of research animals.
c. Safety precautions are necessary to prevent injury and exposure.
d. Identify sharps devices to be used in laboratory procedures. When possible, substitute a non-sharp alternative such as a blunt needle or a plastic pipette, or consider using a safe sharp device.
e. Training and practice are essential to prevent injury. Avoid factors and conditions that can lead to a sharp injury, such as hurrying or rushing or working when you are tired or not feeling well.
f. Keep the working area organised and uncrowded so that sharp items are always visible.

g. Avoid recapping needles. If a needle needs to be recapped, use a needle holder to do so. Never leave an uncapped needle exposed in the work area.

h. Store reusable sharp items in a labelled storage container such as a bucket or tray. Use a magnet to contain reusable metal sharp items like razor blades.

i. Promptly place all sharps waste into a red sharps container. The container must be decontaminated by autoclaving before discarding.

j. When working with needles: -

   i. Use extreme caution to avoid accidental injection and generation of aerosols during use and disposal.

   ii. Use needles only for injection and aspiration of fluid from laboratory animals and diaphragm bottles.

   iii. Do not use a syringe and needle as a substitute for a pipette when making dilutions of fluids. Syringe-type pipettes with blunt-ended delivery are permissible.

   iv. Use needle locking syringes or disposable syringe-needle units in which the needle is an integral part of the syringe.

   v. Prior to beginning an animal inoculation, be sure the animal is properly restrained. Swab the site of the injection with a suitable disinfectant. Inoculate the animal with a hand behind the needle to avoid punctures. Swab the injection site again with a suitable disinfectant.

   vi. Needles should not be bent, sheared, or removed from the syringe for the following use. If you need to change the needle after drawing up a dose, use a tool to remove it.

k. Sharp Items Management: -

   In the laboratory, a sharp item refers to any object that is contaminated with a biologically hazardous agent and is sharp enough to puncture the skin without excessive applied pressure or force. While needles and scalpels could be considered the most apparent objects in this category, other items may also be applied to this definition. Some examples include:

   i. Broken glassware.

   ii. Serological pipettes (especially if broken or damaged).

   iii. Pasteur pipettes.

   iv. Metal edges.

   v. Unpolished glass (slides and cover slips).

The penetration of the skin with a biologically contaminated sharp device is one of the most efficient means of transmitting infection. To minimise the risk of a sharp item injury, the following guidelines must be followed: -

i. Use a disposable sharp item when possible and have the sharps container readily available within arm's reach for the disposal of sharps immediately after use (do not manipulate (recap) by hand before the disposal).
ii. Needles must not be bent, sheared, broken, recapped and removed from disposable syringes, or otherwise manipulated by hand before the disposal.
iii. Do not pass a sharp item to another person.
iv. When cleaning and processing reusable sharps, use cleaning tools that limit the potential for contact between your hands and the sharp surface.
v. Do not leave sharps unattended.
vi. Do not leave sharp devices in your pockets.
vii. Broken glassware is a risk of injury in BSL-1 labs and a risk of injury and infection in BSL-2 labs. It is prudent to substitute plastic for glass in BSL-2 labs whenever possible. If you have large biologically contaminated broken glass items, they must be treated as sharps. Always wear gloves and use tongs or a brush and dustpan to collect broken glassware.

l. Recapping Needles: -

i. Needles are not to be recapped at BSL-2 unless it is specifically exempted by IBC committee. Needle recapping is discouraged at BSL-1, but if necessary, the one-handed scoop technique may be used.
ii. First, place the cap in a flat surface. Second, using one hand, scoop the cap up with the needle. Third, using the same hand, "click" the cap in place by pressing with your thumb at the base of the needle cap.

First, place cap on a level horizontal surface; gently slide needle half-way into cap.

Then, slowly tip up needle end of the device and allow cap to slide over needle

Finally , use the thumb of the hand holding the device to secure the cap on the syringe

(Image from Safe Operating Procedure: SHARPS – HANDLING AND DISPOSING, University of Nebraska Lincoln)

Figure 3.2: Needle recapping process

3.4.3 Centrifugation

a. Accidents resulting from the improper use of centrifuges are rare; however, if accidents do occur, aerosols can be created, which increases the possibility of causing lab-acquired infections.
b. Even a well-functioning centrifuge is capable of producing biohazardous aerosols.
c. Aerosols can be avoided by observing sound laboratory practices.
d. Properly maintain centrifuges according to the manufacturer's instructions to reduce the risk of mechanical failures. For centrifugation of biohazardous agents:
    i. Use a centrifuge with an aerosol containment device such as sealed safety cups or a sealed rotor. Remove the aerosol containment device and open it inside of a BSC.
    ii. If a centrifuge with the above controls is not available, place and operate the centrifuge within a BSC.
    iii. Contact a BSO for assistance with alternative controls or practices if neither option is possible.
e. The greatest hazard associated with centrifuging biohazards is when a centrifuge tube breaks.
f. Do not use glass centrifuge tubes for biohazardous materials. Instead, use plastic tubes and bottles because they resist breakage.
g. Be aware of signs of deterioration in plastic such as crazing, cracking or spotting. Check to see if plastics are compatible with chemical components to be centrifuged. If not, choose a different container.

h. Do not fill tubes to the point that the rim of the closure becomes wet with culture. Pay special attention when filling tubes to be placed in a fixed-angle centrifuge.
i. Do not fill tubes so high that the liquid can spill out when the tube is at an angle.
j. Never use aluminium foil or cotton plugs to cap centrifuge tubes containing biohazards because they can detach or rupture.
k. Instead, use tight-fitting tabbed or hinged caps made of plastic or rubber, screw caps, or other tight-fitting plastic or metal closures.
l. Use screw caps or other tight-fitting skirted caps that fit outside the rim of the centrifuge tube as it is safer than plug-in closures.
m. Some fluid usually accumulates between the plug-in closure and the rim of the tube. Even screw-capped tubes and bottles are not without risk; if the rim is soiled and seals imperfectly, some fluid will escape down the outside of the tubes.
n. Proper balancing of the centrifuge is essential. Extra caution is necessary to ensure that matched sets of safety devices and adapters do not become mixed.
o. If the components are not inscribed with their weights by the manufacturer, put a label to avoid confusion.
p. Properly maintain centrifuges according to the manufacturer’s instructions to reduce the risk of mechanical failures.
q. Follow the manufacturers' recommendations for cleaning and disinfection of the tubes, aerosol containment devices, rotors, and other centrifuge components.
r. Periodically inspect all components, including the sealing gaskets, for any wear out. When problems are noted, the components must be replaced.
s. If there is any centrifuge malfunction and/or spill that may create hazardous aerosols, vacate that room for at least 30 minutes to allow the aerosols to dissipate.
t. Contaminated areas, broken glass, etc., should then be properly decontaminated and cleaned up promptly.
u. The person using the centrifuge, together with the PI and/or laboratory manager, are responsible for ensuring that clean-up and decontamination are achieved. Maintenance service may be refused on centrifuges that appear to be improperly used and/or contaminated.
v. When centrifuging Infectious Materials or Human Samples, follow the safety procedures, plus:
    i. Place a biohazard label on the centrifuge.
    ii. Always wear gloves when handling tubes or rotors.

iii. Avoid the use of celluloid tubes with biohazards. If celluloid tubes must be used, an appropriate disinfectant must be applied to decontaminate them.

iv. Always use sealed safety cups, safety buckets, or sealed rotors with O-ring as secondary containment if available.

v. Fill centrifuge tubes, load into rotors, remove from rotors, and open tubes within a biological safety cabinet if biological safety cabinet is available.

vi. Wipe the exterior of tubes or bottles with disinfectant before loading into the rotor or bucket. Seal rotor or bucket, remove outer gloves, and transport to the centrifuge.

vii. If possible, wait approximately 10 minutes after the run to allow aerosols to settle before opening the centrifuge. Check for possible spills or leaks. For spills of infectious materials, see centrifuge emergency procedures below.

viii. Decontaminate centrifuge interior, safety cups or buckets, and rotors if tube breakage occurs.

ix. Include centrifugation procedure and decontamination plan in laboratory SOPs.

w. Centrifuge Emergency Procedures:-

i. Emergency Situations.

The following events are considered an emergency:

- If there is a spill in the centrifuge.
- If centrifuge malfunctions.
- If there is a rotor failure.
- If there is a tube breakage.

ii. Emergency Procedures.

For emergencies with RG-2 biological agents: -

- Turn centrifuge off immediately and keep centrifuge lid closed.
- Notify others.
- Evacuate the laboratory if necessary.
- Close the laboratory door.
- Post a biohazard spill sign at the laboratory door.
- Leave for 30-minutes to reduce the risk of aerosols.

- For spill clean-up, the operator should wear proper PPE, remove debris, clean and disinfect the centrifuge interior, rotors, safety cups or buckets following the manufacturer's instructions.
- Place any contaminated protective clothing, gloves, and all cleaning materials in a biohazard bag.
- Wash hands and any exposed skin surfaces with soap and water.
- Seek medical attention as necessary.
- Report incidents to PI or laboratory manager and Biosafety Officer if the incident involves chemical hazards.

3.4.4 Re-suspending Sediment of Centrifuged Material

a. Use a swirling, rotary motion rather than shaking to suspend the sediment of packed biohazardous materials.
b. This motion minimises the amount of aerosol created.
c. Perform these operations inside a BSC. If vigorous shaking is essential to suspend the material or achieve homogeneity, allow a few minutes to elapse before opening the container to allow the aerosol to settle.
d. Shaking always contaminates the closure and creates the added hazard of liquid escaping and running down the outside of the container or dropping from the closure when it is removed.

3.4.5 Opening Culture Plates, Tubes, Bottles, and Ampoules

a. Aerosol formation is the primary concern when plugs or screw caps are removed from tubes and bottles. Slow and smooth manipulations will minimise the formation of aerosols.
b. Opening ampoules is potentially hazardous since after the seal is broken, the air rushes in causing the dry contents to be dispersed.
c. A BSC should be used.
d. The bottom of the ampoule should be held in several layers of laboratory wipes to protect the hands. Scratch the neck of the ampoule with a file. A hot glass rod should be carefully applied to the mark.

e. The glass will crack, allowing air to enter the ampoule and equalise the pressure. After a few seconds, the ampoule should be wrapped in a few layers of laboratory wipes and broken along the crack. An alternative method of opening an ampoule involves wearing gloves and other PPE, nicking the ampoule with a file, and wrapping the ampoule in disinfectant wetted cotton for breaking.
f. The ampoule neck and other waste are handled as biohazardous sharps waste in both methods.

3.4.6 Using Test Tubes and Other Laboratory Glassware

a. Tubes containing biohazards should be manipulated with extreme care.
b. Studies have shown that simple procedures such as removing the tube cap or transferring an inoculant can create a potentially hazardous aerosol.
c. Tubes and racks of tubes containing biohazards should be clearly marked with agent identification.
d. Safety test tube trays should be used in place of conventional test tube racks to minimise spillage from broken tubes. A safety test tube tray has a solid bottom and sides that are deep enough to hold all liquids if a tube should break.
e. Glassware breakage is a major risk for puncture infections. Avoid unnecessary use of glass Pasteur pipettes. Whenever possible, use flexible plastic pipettes or other alternatives.
f. It is the responsibility of the PI and/or laboratory manager to ensure that all glassware/plasticware is properly decontaminated prior to washing or disposal.

3.4.7 Transporting

Transferring or transporting infectious substances within or between laboratories should always be undertaken in a way that minimises the potential for drop, spillage, collision or similar events. The following subsections provide an overview of the main issues to consider in transferring or transporting an infectious substance.

a. Transfer within the laboratory.
    i. Use sealed containers, such as screw-capped tubes. Snap-cap lids should be avoided, as they are less secure.

ii. Use deep-sided and leak-proof trays or boxes made of smooth impervious material (e.g. plastic or metal), which can be effectively cleaned and disinfected. Locking plastic food storage containers and storage containers are an option.

iii. If using racks, vials or tubes, trolleys can be used for more stable transport, as they are less likely to result in multiple spillages if a worker trips or falls.

iv. If using trolleys, ensure they are loaded so that substances cannot fall off, e.g. by securing the load or using some form of guard rail or raised sides.

v. Make sure spill kits are readily available for use in the event of spillage during transfer, and available personnel are trained in their use.

b. Transfer within a building.

i. In addition to the considerations above, the transfer of infectious substances between rooms, departments or laboratories in the same building must be planned, organised and carried out in a way that minimises transit through communal areas and public thoroughfares.

ii. Transfer containers must be suitably labelled to identify their contents and surfaces decontaminated before leaving the laboratory. Biohazard symbols should be used on containers as a heightened control measure if the biological agent being handled is associated with a higher likelihood of infection.

c. Transfer between buildings on the same site.

Issues that need to be considered for containers and layers of outer packaging to minimise the risks of leakage while transferring infectious substances between buildings are outlined below: -

i. Sealable plastic bags, plastic screw top tubes and locking plastic food storage containers.

ii. Absorbent materials should be used between the layers of packaging to absorb all infectious substances in the event of a leakage.

iii. The outermost transport container should be rigid. It can vary widely depending on the resources available. A plastic lunch box or small plastic ice chest (Figure 9) is one option for transporting infectious substances between buildings on the same site, as it is secured and easily decontaminated.

iv. Packaging should be labelled in a way that the sender, recipient and contents of the package are clearly identifiable. It should include biohazard symbols where appropriate.

v. Personnel involved in the transfer must be provided with suitable awareness training on the risks present during the transfer process and how to reduce them safely.
vi. Spill kits must be readily available and appropriate personnel trained in their use.
vii. Recipients must be notified in advance of the transfer occurring.

3.5 DECONTAMINATION

Decontamination is a process or treatment that reduces biohazardous material, which renders a medical device, instrument or environmental surface safe to handle to an acceptable level.

3.5.1 Factors that Affect Decontamination

a. Resistance
   i. Microorganisms exhibit a range of resistance to chemical decontaminants. Most vegetative bacteria, fungi and lipid-containing viruses are relatively susceptible to chemical decontamination.
   ii. The non-lipid containing viruses and bacteria with a waxy coating occupies a mid-range resistance.
   iii. Bacterial spores are the most resistant microorganism. The relative resistance to the action of chemical decontaminants can be substantially altered by a few factors such as concentration of an active ingredient, contact time, pH, temperature, humidity, and presence of extrinsic organic matter.
   iv. Depending on how these factors are manipulated, the degree of success achieved with chemical decontaminants may range from minimal inactivation of target microorganisms to an indicated sterility within the sensitivity limits of the assay systems employed.

b. Ineffectiveness
   i. The ineffectiveness of a decontaminant is primarily due to the failure to contact the microorganisms rather than the failure of the decontaminant's action.
   ii. If an item is placed in a liquid decontaminant, the item becomes covered with tiny bubbles. The area under the bubbles is dry and microorganisms in these dry areas will not be affected by the decontaminant.

iii. If there are spots of grease, rust or dirt on an object, microorganisms under these protective coatings will also not be contacted by the decontaminant.

c. Residual Action

i. Many chemical decontaminants have residual properties that may be considered a desirable feature in aiding in controlling background contamination. However, residual properties need to be considered carefully.

ii. Ethylene oxide can leave residues that may cause skin irritation. Concentrated phenol readily penetrates the skin and causes severe burns.

iii. Animal cell cultures and viruses of interest are also inhibited or inactivated by decontaminants persisting after routine cleaning procedures.

iv. Therefore, reusable items that are normally held in liquid decontaminants prior to autoclaving and cleaning require careful selection of detergents for washing and must be thoroughly rinsed.

d. Exposure Time

i. Specific exposure times for the decontamination process with autoclave, dry heat or chemical decontaminants cannot be determined.

ii. The volume of material treated, its contamination level, moisture content and other factors play a role in the inactivation rate of microorganisms.

3.5.2 Levels of Decontamination

The levels of decontamination range from high-level sterilization to simple cleaning with soap and water. The levels of decontamination are listed as follows:

a. Sterilization

Sterilization uses a physical or chemical method that aims to eliminate or destroy all microbial life including highly resistant bacterial endospores.

b. Disinfection

i. Disinfection uses a liquid chemical to eliminate all pathogenic microorganisms with the exception of bacterial spores on work surfaces and equipment.
ii. The effectiveness of disinfection depends on the kind and number of organisms, the amount of organic matter, the object that needs to be disinfected and the exposure time, temperature and concentration of the liquid chemical used.

c. Antisepsis
   i. Antisepsis is the application of liquid antimicrobial chemicals to skin or living tissue to inhibit or destroy microorganisms.
   ii. Antisepsis includes swabbing an injection site on a person or animal and hand washing it with germicidal solutions.

d. Cleaning
   i. Water, detergent and mechanical action such as scrubbing with a gloved hand or brush are used in the cleaning method.
   ii. It is considered as a required step before sterilisation or disinfection of inanimate objects because materials such as soil or organic material are removed, and the number of microorganisms on an object is reduced.

3.5.3 Type of Decontamination

There are FOUR types of physical and chemical decontamination methods, which are heat sterilisation, liquid disinfection, vapours and gases and radiation. Each method is described below.

a. **Heat Sterilisation**
   i. **Wet Heat**
      - Wet heat is also known as autoclaving, the most convenient and dependable method to achieve effective and rapid sterilisation of most forms of microbial life.
      - Autoclaving uses the application of saturated steam under pressure approximately to achieve a chamber temperature of 120°C for periods of 30-60 minutes. Wet heat is more efficient than dry heat due to the shorter time and lower temperature required.

ii. **Dry Heat**

- Dry heat is used under the conditions of 160-170 °C for 2 -4 hours in an appropriate oven.
- An impermeable non-organic surface such as glass and other non-porous heat conductive materials are normally sterilised by dry heat. However, this method is not suitable for insulation or heat-labile materials.

b. **Liquid Disinfection**

Liquid disinfectants are widely used for laboratory decontamination. The effectiveness of liquid disinfectants depends on several factors such as the chemical nature of the disinfectant, concentration and quantity of disinfectant, contact time and temperature. **Table 3.3** shows the types of liquid disinfectants that are generally used for laboratory decontamination purposes.

Suitable liquid disinfectants should be selected according to the detected microorganisms. Criteria for the selection of suitable liquid disinfectants are as follow:

i. Type of contaminating microorganism.
ii. Amount of proteinaceous material present.
iii. Amount of organic material present.
iv. Other important factors include the chemical nature, concentration, quantity, pH, application temperature and toxicity of disinfectant.

Table 3.3: List of liquid disinfectants* used for decontamination

Type of Chemicals	Advantages	Disadvantages
Quaternary Ammonium (QA) compounds Example: Benzalkonium chloride and Ammonium chloride	• Effective against Gram-positive and Gram-negative bacteria and enveloped viruses • Contain $NH^{4+,}$ which can provide good contact with negatively charged surfaces	• Not effective against non-enveloped viruses, fungi and bacterial spores
Phenolic	• Effective against Gram-positive bacteria and enveloped viruses	• Not effective against non-enveloped viruses and spores

Example: O-phenophenoate based compounds	• Compatible with organic materials • Low in toxicity	• Irritate when exposed for long durations
Alcohols Example: Ethyl alcohol and isopropyl alcohol	• Effective against Gram-positive and Gram-negative bacteria and enveloped viruses • The optimum concentration is between 60 and 90%	• Not effective against spores • Limited effect against non-enveloped viruses • Activity decreases when the concentration is below 50% • Flammable and easily evaporate
Chlorine-based compounds Example: Hypochlorite	• Effective against both enveloped and non-enveloped viruses, fungi, bacteria and algae	• Not effective against spores • Easily inactivated by organic matter • Due to high oxidising power, it can be degraded quickly
Iodophors	• Effective against bacteria, spores and fungi	• Require prolonged contact time • Not effective in the presence of organic matter
Oxidiser Example: Hydrogen peroxide	• Effective against enveloped and non-enveloped viruses, vegetative bacteria, fungi and bacterial spores	• High concentration of hydrogen peroxide is harmful to tissue

Acid Example: Peracetic acid	• Effective against all microorganisms with rapid action • Effective in the presence of organic matter and low temperature • It is safe with no harmful decomposition products	• Not suitable for metal surface due to corrosion
Aldehyde Example: Formaldehyde and Glutaraldehyde	Formaldehyde • Effective against bacteria, fungi, viruses and spores Glutaraldehyde • Effective against vegetative bacteria, spores and viruses • Effective in the presence of organic material	Formaldehyde • Hazardous with eight hours' time-weighted exposure limit of 0.75 ppm Glutaraldehyde • Hazardous with ceiling threshold limit 0.2 ppm

*The correct sequence for successful disinfection is: (i) clean the surface to remove dirt and organic matter, (ii) apply the disinfectant and (iii) after a pre-determined contact time, wipe with water to remove chemical residues, if necessary.

Generally, higher concentrations of highly-active disinfectants are required to inactivate spores than enveloped viruses. Generally, a solution of sodium hypochlorite (NaOCl) with 1000 parts per million (ppm) is used for general surface disinfection, in which the existing chlorine will suffice. However, stronger solutions (for example, 5000 ppm or 10 000 ppm) will be required when dealing with heavy contamination or the presence of organic matter or disinfectant-resistant biological agents.

Hypochlorites, for example, are inactivated by organic substances. Higher concentrations are required even at relatively low levels of contamination ("clean conditions"), and the concentration required increases as the amount of organic matter contamination increases ("dirty

conditions"); see Table 3.5. Hypochlorite solutions can be produced from a mixture of starting agents, including liquid bleach. This is a low-cost option that is readily available. However, because liquid hypochlorite decay during storage, the available chlorine concentration of these solutions might be imprecise. The rate of decay is dependent on storage conditions, particularly temperature. The shelf life of diluted hypochlorite solutions is limited, usually, one day, depending on exposure to heat and/or sunlight. **Table 3.4** shows the recommended dilutions of compounds releasing chlorine.

Table 3.4: Recommended dilutions of compounds releasing chlorine.

HYPOCHLORITE SOURCE (PERCENTAGE AVAILABLE CHLORINE)	CLEAN CONDITIONS[a] (AVAILABLE CHOLORINE NEEDED FOR DISINFECTION: 1 G/L – 0.1% AVAILABLE CHLORINE – 1000 PPM AV CL)	DIRTY CONDITIONS[b] (AVAILABLE CHOLORINE NEEDED FOR DISINFECTION: 5 G/L – 0.5% AVAILABLE CHLORINE – 5000 PPM AV CL)
Sodium Hypochlorite solutions (5% available chlorine)	20 mL	100mL
Calcium Hypochlorite (70% available chlorine)	1.4 g/L	7.0 g/L
Sodium dichloroisocyanurate powder (60% available chlorine)	1.7 g/L	8.5 g/L
Choloroamine powder (25% available chlorine)	4 g/L	20 g/L

ppm av cl = parts per million available chlorine

[a] Low levels of contamination

[b] High levels of contamination

c. **Vapours and Gases**

i. Vapours and gases provide excellent disinfection properties when used in closed systems with temperature and humidity-controlled conditions.

ii. Vapours and gases are primarily used to decontaminate biosafety cabinets, animal room facilities, bulky or stationary equipment that are not suited to liquid disinfectant and instruments or optics that might be damaged by other decontamination methods.

iii. Agents of this disinfection method include vapour or gas phase chlorine dioxide, glutaraldehyde, paraformaldehyde, ethylene oxide, acetic acid and hydrogen peroxide.

d. **Radiation**

There are two general types of radiation used for decontamination purposes, ionizing and non-ionizing radiation.

i. Ionizing Radiation:

- Ionizing radiation uses short wavelength and high-intensity radiation to destroy microorganism.
- This type of radiation usually comes in the form of gamma rays or X-rays that react with DNA resulting in cell damage. Ionizing radiation is not a practical tool for general laboratory sterilisation due to potential issues associated with radiation safety.

ii. Non-ionizing Radiation:

- Longer wavelength and lower intensity (lower energy) of radiation are used for non-ionizing radiation.
- The most common form of non-ionizing radiation is ultraviolet (UV) radiation. UV with C band (UV-C) is generally used in the laboratory to effectively destroy most of the microorganisms in the air, water and on surfaces. It contains wavelength ranges from 250-270 nm, with 265 nm as the optimum wavelength used for disinfection.
- UV radiation is typically used to reduce levels of airborne microorganisms and maintain good air hygiene in air locks, animal holding areas, ventilated cabinets, and laboratory rooms.
- UV is also used in BSC and in some laboratory rooms to reduce surface contamination. The equipment used for decontamination is called a UV lamp.

Maintenance

Maintenance needs to be performed on the UV lamp in order to maintain the power of UV as its radiation intensity reduces with time. The lamp needs to be checked monthly with a UV meter or monitoring strip and should be cleaned every few weeks to prevent the accumulation of dust and dirt that can reduce the effectiveness of the UV lamp.

Precaution step

UV radiation can cause burns to the eyes and skin, hence precautions need to be taken to ensure the safety of the people. The precautions are as below:

- The UV light can only be activated when the area is unoccupied.
- Proper shielding needs to be installed and used when using the UV lamp.
- Interlocking the UV lamps used for space decontamination with general room or cabinet illumination. By interlocking these two systems, the UV light will be turned off when the lights of the general room or cabinet are turned on.
- Display the hazardous effects of UV radiation and precautions should be taken in a simple and clear view to inform and warn visiting or new personnel.

3.6 BIOHAZARDOUS WASTE

Biohazardous waste, also known as infectious waste or biomedical waste, is any contaminated waste with infectious materials or potentially infectious agents that are considered a threat to public health or the environment.

3.6.1 Identify Biohazardous Waste

Types of Biohazardous Waste:

a. Solid Biohazardous Waste

Solid biohazardous waste is any non-sharp materials contaminated with human or animal specimens such as body fluids and microbiological culture material. Examples of solid biohazardous waste are as follows:

i. Personal Protective Equipment (PPE).
ii. Towels, bench papers, linens, Kim wipes.
iii. Plasticware such as pipettes tips.
iv. Culture or sample containers such as petri dish, inoculum.

b. Sharp Waste

Sharps waste refers to contaminated items that can cause puncture and cut to skin or body parts. Examples of sharp waste are as follows:

i. Needles.
ii. Intravenous **(**IV) tubing with a needle attached.
iii. Scalpels, razor blades and lancets.
iv. Broken glass and splintered plastic contacted infectious agents.
v. Microscopic slides and coverslips.
vi. Glass Pasteur pipettes.

c. Liquid Biohazardous Waste

Human blood, blood products and human body fluids, whether in liquid or semi-liquid form are classified as liquid biohazardous waste. Examples of liquid biohazardous waste are as follows:

i. Blood products: serum, plasma and other blood components.
ii. Body fluids: saliva, semen, vaginal secretions, cerebral spinal fluids, amniotic fluids, synovial fluids, pleural fluids, pericardial fluids and peritoneal fluids.

d. Pathological Waste

Pathological waste is human organs, tissue and body parts which include waste biopsy materials, tissues and anatomical parts from surgery, procedures or autopsy.

e. Animal Waste

Animal waste is animal carcasses, body parts and any bedding material from animals that have been exposed, infected or inoculated with human pathogenic microorganisms that are infectious to humans. (**refer sec.6**)

f. Microbiological Waste and Recombinant DNA and RNA/LMO/GMO

i. Cultures or stocks of pathogenic agents include bacteria, fungi, viruses, protozoa, parasites, prions and selected agents.
ii. This category includes discarded live and attenuated viruses, wastes from the production of biological and serums.
iii. Recombinant or synthetic nucleic acids include waste products from procedures involving plasmids, viral vectors, *E. coli*, yeasts and naked nucleic acids.

3.6.2 Packaging, Handling, Treatment, Labelling and Disposal of Biohazardous Waste

a. Solid Biohazardous Waste

i. Packaging and Handling

- This type of waste should be collected in a collection container lined with a yellow autoclavable biohazardous waste bag and marked with a biohazard symbol.
- The collection container needs to be rigid, leak-proof and must have a lid. The lid and the sides of the container need to be clearly labelled and displayed with a biohazard symbol.
- The maximum storage of solid biohazardous waste is seven days. Once the bag is 2/3 full, tie the bag loosely with a single overhand knot to allow steam to penetrate during autoclaving.
- **Figure 3.3** shows the biohazardous waste collection container and yellow biohazardous waste bag.

ii. Treatment

- Autoclave.

iii. Labelling

- Biohazard symbol must be on the lid and the sides of the collection container.

- Laboratory's address.

iv. Disposal

- Once the bag is 2/3 full, tie the bag loosely with a single overhand knot to allow steam to penetrate during autoclaving.
- The waste bag can be placed in a yellow plastic waste container with a biohazard symbol at the designated biohazardous waste collection site after being autoclave. The waste will be picked up weekly by the scheduled waste contractor assigned by UTHM.


Figure 3.3: The collection container and the yellow plastic bag for solid biohazardous waste

b. Sharp Waste

i. Packaging and Handling

Sharp wastes need to be collected in a yellow plastic sharp container labelled with a biohazard symbol. The size of the container should fit the workspace. The container must be leak-proof, rigid and puncture-resistant. An example of a sharps container is shown in **Figure 3.4**.

ii. Treatment

- Off-site treatment by the scheduled waste contractor assigned by UTHM

iii. Labelling

- Biohazard symbol.
- Laboratory's address.

iv. Disposal

- When the container is 2/3 full, close the lid. Place the sharps container at the designated biohazardous waste collection area. The waste will be picked up weekly by the scheduled waste contractor assigned by UTHM.

Figure 3.4: Sharps container

c. Liquid Biohazardous Waste

i. Packaging and Handling

- The liquid must be stored in a closed and leak-proof container or vacuum flask with a stopper and labelled with a biohazard symbol.
- A collection container needs to be secured to avoid overturning. A secondary container such as a tray is strongly recommended as an additional security measure.

ii. Treatment

- Autoclave.
- Chemical disinfection.
  - The waste should be collected in a vacuum flask with a biohazard symbol.
  - The bleach will be added to the vacuum flask containing waste with a ratio of 9:1 (9 parts of waste, 1 part of bleach).
  - The vacuum flask is then left at room temperature for 30 minutes to ensure sufficient contact time with disinfectant.

iii. Labelling

- Biohazard symbol

- Label flask to identify liquid biohazardous waste and bleach (9:1).

iv. Disposal

Most liquid biohazardous waste can be disposed of through a laboratory sink using appropriate PPE after chemical treatment (liquid disinfection) or autoclaving. The sink must be rinsed with plenty of water.

d. Pathological Waste

i. Packaging and Handling

- The pathological waste must be placed in a yellow plastic bag for solid biohazardous waste and tie the bag tightly**.**

ii. Treatment

- Off-site treatment by the scheduled waste contractor assigned by UTHM

iii. Labelling

- Biohazard symbol
- Pathological waste or PATH
- Laboratory's address

iv. Disposal

- The bag needs to be tied closely before transferring to the designated biohazardous waste collection area. The waste will be picked up weekly by the scheduled waste contractor assigned by UTHM.

e. Microbiological waste and Recombinant DNA and RNA

i. Packaging and Handling

- The waste must be stored in a closed and leak-proof container or vacuum flask with a stopper and labelled with a biohazard symbol.
- A collection container needs to be secured to avoid overturning. A secondary container such as a tray is strongly recommended as an additional security measure.

ii. Treatment

- Autoclave
- Chemical disinfection
  - The waste should be collected in a vacuum flask with a biohazard symbol.
  - The bleach will be added to the vacuum flask containing waste with a ratio of 9:1 (9 parts of waste, 1 part of bleach).
  - The vacuum flask is then left at room temperature for 30 minutes to ensure sufficient contact time with disinfectant.

iii. Labelling

- Biohazard symbol
- Label flask to identify microbiological waste and bleach (9:1). An example of this label is shown in **Figure 3.5**.

Figure 3.5: Example of biohazard label with waste and bleach

iv. Disposal

- Most liquid biohazardous waste can be disposed of through a laboratory sink using appropriate PPE after chemical treatment (liquid disinfection) or autoclaving.
- The sink must be rinsed with plenty of water.

3.6.3 Transporting Biohazardous Waste

This section describes the correct procedure for transporting biohazardous waste within buildings and in between buildings. Biohazardous waste must be packaged so that PPE usage is not needed during transportation.

a. Within Building

i. Sharps Containers

Sharps containers that are transported within the same building must be securely closed and the outer surface must be decontaminated prior to the transportation process. It is recommended to place the sharps containers inside a secondary container to avoid leakage. The secondary container must be closable, puncture-resistant and leak-proof. This container should be labelled with a biohazard symbol.

ii. Other Biohazardous Waste

Bagged biohazardous waste transported within the same building must be closed, surface decontaminated and placed inside a secondary container prior to transport. It is not allowed to transport biohazardous waste in biohazard bags alone. If the secondary container has a lid or closure, it must be identifiable as biohazardous either by being yellow in colour or labelled with a biohazard symbol.

b. Between Buildings

Any biohazardous waste that is transported between buildings by motor vehicle must be transported using an institution-owned vehicle. Sharps containers that need to be transported between buildings must have the same requirements as within the building. Bagged biohazardous waste transported between buildings has the same requirements as within the same building with the exception that the secondary container must have a secured lid.

SECTION 4: EMERGENCY PREPAREDNESS AND RESPONSE PLAN

An emergency response plan (ERP) is a series of documents for dealing with laboratory incidents and accidents which is a necessity in any facility that works with or stores biological hazards. The head of the centres should implement the ERP and establish a response team (ERT) in case of a biological emergency.

4.1 BIOHAZARDOUS SPILLAGE

Good Laboratory Practice (GLP) embodies a set of principles that provides a framework within which laboratory studies are planned, performed, monitored, recorded, reported and archived.
Biohazardous spillages come in two types:

a. Unintended release of potentially infectious material which has been handled or generated within a laboratory (e.g.: liquid cultures of micro-organisms).
b. Human or animal bodily fluids or materials such as blood, urine, vomit or faeces either accidentally or maliciously.

A proper response to such incidents ensures the safety of employees and students while reducing the environmental contamination concerns. The responses include assuring the spillage kits are available for use and verifying that all personnel understand and are able to implement the requirements of the spill response procedures shown in the latter part of this section.

4.2 SPILL RESPONSE PROCEDURES

4.2.1 Low-Risk Spills

a. The following protocol is for low-risk biohazard spills of low-risk materials such as microbiological cultures of known origin and risk and for spills of blood or body fluids.
b. Notify others that are working in the hazard area and put up signage (e.g., “Biohazard Spill – Do Not Enter”) if necessary.
c. Notify your supervisor and IBC so they can supervise and/or assist with spill response if needed.
d. Note if your clothing or skin comes in contact with the spilled material, take appropriate action before proceeding with clean-up. Remove contaminated clothing pieces and place them in a

designated biohazard bag for decontamination. Thoroughly wash the area of your skin that may have been in contact with the spilled material with soap and water for about five minutes.

e. Wear gloves, eye protection, and a protective lab coat as minimum personal protective equipment (PPE). Replace PPE that is damaged or becomes contaminated before proceeding with clean-up.
f. If applicable, use tongs or forceps to pick up any contaminated sharp items (syringes, broken glass, etc.) and place them in the sharps container for disposal.
g. Put an absorbent material, such as a paper towel, over the spill.
h. Isolate the spilled material starting with the outer edges of the towel and working into the centre of the spill. Soak the area with 10% household bleach solution (prepared as needed for maximum effectiveness). Note: An EPA registered disinfectant can be used as a substitute for bleach.
i. Allow the treated towels to stand for a minimum of five minutes. Then, collect the treated towels using scoops or other mechanical methods and place them in the biohazard bag for disposal.
j. Wipe the entire spill, including articles in close proximity that may not be visibly contaminated, with clean towels and bleach/disinfectant solution. Allow it to air dry.
k. Place reusable spill response items in the autoclave (biohazard) bag; sterilize items prior to further processing for reuse if items can tolerate the heat of the autoclave; if not, soak in disinfectant for > 10 minutes, then clean before reuse.
l. Place all disposable PPE and cleaning materials in another biohazard bag to be treated and disposed of. Autoclave the bag and enclose the treated waste in a regular trash bag prior to disposal.
m. If you need help, contact the IBC personnel.

4.2.2 Spills outside Laboratory in Public Spaces

a. Samples must be transported in secondary, leak-proof containers to minimise the potential for spills.
b. However, if a spill does occur in a common hallway or public space and cannot be immediately decontaminated, cordon off the area, restrict access and contact the IBC personnel.
c. Be sure to stay at the spill location until help arrives.

4.2.3 Spill outside Laboratory during Transport on Campus

a. Always transport biohazardous materials in an unbreakable well-sealed primary container placed inside a leak-proof, closed and unbreakable secondary container, labelled with the biohazard symbol (plastic cooler, bio-specimen pack, etc.).
b. Should a spill of biohazardous material occur in public, directly contact the IBC personnel. Do not attempt to clean up the spill without proper personal protective equipment and spill clean-up materials.
c. Should the spill occur inside a car, leave the vehicle, close all doors and windows, and contact IBC personnel for assistance.

4.2.4 Spill inside Laboratory

a. Clear the spill area. Wait for the aerosols to settle before entering the spill area.
b. Remove any contaminated clothing and place it in a biohazard bag for further processing by laundry.
c. Wear a disposal gown or lab coat, safety goggles and gloves.
d. Have a complete biohazardous kit ready to go before you start the clean-up.
e. Initiate clean-up with disinfectant as follows:
    i. Cover spill with paper towels or other absorbent material containing disinfectant.
    ii. Encircle the spill with disinfectant (if feasible and necessary), being careful to minimise aerosolisation.
    iii. Decontaminate and remove all items within the spill area.
    iv. Remove broken glassware with forceps or broom and dustpan and dispose it in sharps container. Do not pick up any contaminated sharp object with your hands.
    v. Remove paper towels and any other absorbent material and dispose it in biohazard bags.
    vi. Apply the disinfectant to the spill area and allow for at least 10 minutes contact time to ensure germicidal action of disinfectant.
    vii. Remove disinfectant with paper towels or other absorbent material and dispose of in biohazard bag.
    viii. Wipe off any residual spilled material and reapply disinfectant before final clean-up.
    ix. Wipe the equipment with compatible disinfectant (e.g.: non-corrosive). Rinse with water if necessary.
    x. Place disposable contaminated spill materials in biohazard bags for autoclaving.

xi. Place contaminated reusable items in biohazard bags, heat resistant pans or containers with lids before autoclaving.

xii. Re-open area for general use only after the spill clean-up and decontamination is complete.

xiii. Inform all personnel and laboratory supervisor about the spill and successful clean-up as soon as possible.

4.2.5 Spill inside Centrifuge

a. Have a complete biohazardous spill kit ready to go before you start the clean-up.
b. Clear the area. Wait 30 minutes for aerosol to settle before attempting to clean up the spill.
c. Wear a lab coat, safety goggles and gloves during the clean-up.
d. Remove rotors and buckets to the nearest biological safety cabinet.
e. Thoroughly disinfect inside the centrifuge.
f. Remove contaminated debris after disinfection, place it in the appropriate biohazardous waste container(s) and autoclave before disposal.

4.2.6 Spill inside Biological Safety Cabinet (BSC)

a. Have a complete biohazardous spill kits ready to go before you start the clean-up.
b. Wear lab coat, safety goggles and gloves during clean-up.
c. Allow cabinet to run during clean-up.
d. Soak up spilled material with disposable paper towels (work surface and drain basin) and apply disinfectant with a minimum of 10 minutes contact time.
e. Wipe up spillage and disinfectant with disposable paper towels.
f. Wipe the walls, work surface and any equipment in the cabinet with a disinfectant-soaked paper towel.
g. Discard contaminated disposable materials in biohazard bag(s) and autoclave it before discarding as waste.
h. Place contaminated reusable items in biohazard bags, heat resistant pans or containers with lids before autoclaving and further clean-up.
i. Expose non-autoclavable materials to disinfectant, 10 minutes contact time, before discarding as waste.

j. Remove protective clothing used during the clean-up and place it in a biohazard bag for further processing by laundry.
k. Run cabinet at least 10 minutes after clean-up and before resuming work.
l. Inform all users of the BSC as well as the laboratory supervisor about the spill and successful clean-up as soon as possible.

4.2.7 Combined Hazard Spill (Radioactivity, Carcinogen)

a. In laboratories, a spill of potentially infectious material may also have other hazardous characteristics.
b. A common additional hazard is radioactivity due to the widespread use of isotopes markers.
c. Procedures that need to be followed are:
    i. Evacuate the area and notify the IBC personnel for response.
    ii. Do not initiate any clean-up activities on your own before radiation safety personnel arrive.
    iii. Radiation safety personnel will survey the affected area to determine the appropriate method of treatment and the disposal of the spill materials. Be prepared to assist responders if needed.
    iv. Following the spill response activities, radiation safety responders will again survey the area to verify that radiation decontamination has been achieved.
    v. Make sure that all personnel stay out of the area until the responders have determined that it is safe to re-enter.

4.3 CHEMICAL DISINFECTANTS

When using a chemical disinfectant, remember that you are using a potentially toxic chemical that could be corrosive, flammable, an irritant, and or potentially carcinogen. Disinfectants must be used according to the product label and be sure to wear the PPE as indicated on the product label and Safety Data Sheet (SDS).

4.4 BIOHAZARDOUS SPILLAGE KITS

Biohazardous Spillage Kits must contain: -

a. An appropriate chemical disinfectant (e.g.: a freshly prepared 1:10 dilution of household bleach, or other decontaminant appropriate for biohazardous agent in use).
b. Absorbent material (e.g.: paper towels, absorbent laboratory pads, or any other special materials designed to absorb large volumes of liquid).
c. Waste container (e.g.: biohazard bags, sharps containers).
d. Personal Protective Equipment (PPE) (e.g.: long sleeve lab coat or gown, nitrile or heavy-duty gloves, safety glasses or goggles and facial protection for large spills, and any additional PPE required for biohazardous agent).
e. Mechanical tools (e.g.: forceps, tongs, scoops, sponges, broom, autoclavable dustpan, or any other method that prevents direct contact with broken glass).

4.5 ACCIDENT REPORTING

a. All spills, accidents and incidents must be reported immediately (24 hours) to the laboratory supervisor and/or IBC using the Notification of Accident, Dangerous Occurrence, Occupational Poisoning, and Occupational Disease (NADOPOD) form (UTHM/OSHE/UP.002) (see Appendix A) and submitted to OSHE UTHM.

b. The injured person should seek medical attention at the nearest medical centre within two hours and, if possible, bring their immunization records with them, any applicable SDS and site-specific Post Exposure Protocol (PEP).

c. Such reporting enables appropriate investigation and follow-up to prevent similar events or more severe incidents in the future. Additional reporting includes:
   i) Where the incident occurred
   ii) For assistance with this process, including determining employee/student status, contact IBC and OSHE UTHM.

d. It is also important to report any near-miss incident, unplanned or unanticipated events where nobody was injured, or nothing was spilt or damaged but only by good fortune. These must also be investigated to prevent the set of circumstances leasing to the event from arising again.

SECTION 5: LMO/GMO EXPERIMENTATION

In UTHM, IBC UTHM acknowledges that modern biotechnology can cause harm to human health and the environment (risk assessment). These risks can be managed or mitigated to a minimal level (risk management/ mitigation). The following is the checklist that principle investigators (PIs) need to follow if they are working with modern biotechnology research:

a. Be aware that your work may have risks.
b. Identify the potential risks.
c. Do a risk assessment – are the risks acceptable?
d. Do risk management/mitigation.
e. Is the risk reduced to an acceptable level?
f. Prepare Emergency Response Plans in case something bad happens.

5.1 BIOSAFETY CHECKLIST

a. Before carrying out the research, PI needs to identify the types of organisms that they are going to use; non-exempted GMO/LMO or exempted GMO/LMO or LO.
b. PI is responsible for ensuring that adequate and timely risk assessments are performed and working closely with IBCUTHM to ensure that appropriate equipment and facilities are available to support the work.

5.2 RISK ASSESSMENT AND RISK MANAGEMENT

Based on the information ascertained during the risk assessment, a biosafety level can be assigned to the planned work, appropriate personal protective equipment selected, and standard operating procedures (SOPs) incorporating other safety interventions developed to ensure the safest possible conduct of the work.

a. PI is required to evaluate the level of risks and fill out a Google form, '*NOTIFICATION FOR ACTIVITIES INVOLVING LIVING ORGANISM (LO) AND LIVING MODIFIED ORGANISM (LMO) IN THE UNIVERSITY'* or manually download the form (UTHM / OSHE / UBK.001

REV.4). Both forms can be found at https://oshe.uthm.edu.my/v3/. The example of the form is shown in **APPENDIX B**.

b. IBCUTHM will ensure that general SOP (*including SOP for every single activity involving work with infectious and potentially infectious agents/materials and microbial toxins, for example; donning and doffing; collection, movement, transport and handling of infectious agents; receiving and storage of infectious agents; proper use of laboratory instruments and equipment; hand washing; entry and exit; disinfection, decontamination and sterilization; spill and waste management; accidents and incidents including loss, theft; and emergency response plan*), as well as agent-specific SOP, are established. (Use SOP form as in **APPENDIX C**).

c. Ideally SOPs:
   i. should be written based on actual activity performed.
   ii. should be written in language that is understood well by the performer.
   iii. is available and easily accessible to all laboratory personnel.
   iv. appropriate and relevant SOP is used as the basis for personnel training.
   v. is evaluated, validated, communicated, periodically reviewed and updated, and documented by the IBC UTHM based on the most acceptable recent reference guidelines.

d. Should there be situations when the information is insufficient to perform an appropriate risk assessment, for example, with unknown biological samples collected in the field, it is prudent to follow the sample-handling procedure as described in the Laboratory Biosafety Manual introduced by World Health Organisation, 2004.

5.3 NON-EXEMPTED USE OF LMO / GMO ACTIVITIES

a. Researchers must fill out and complete **Form E (NBB / N / CU / 15 / FORM E)** which can be downloaded from the NBB's website.
b. The completed Form E must be sent to the Biosafety Unit OSHE for review.
c. IBC assessment will be made by BSO based on the application made.
d. Form E, together with the IBC Assessment Report (**BC / AP / 10 / ANNEX2**) by BSO will be brought to the IBC Chairman for approval.

e. Applications that the IBC Chairman has approved will be forwarded to NBB through Biosafety Unit OSHE for approval.
f. Acceptance of Notification Receipt will be issued for each application received by NBB.
g. Researchers must provide additional information related to the application if requested by NBB.
h. NBB will issue the decision on the application within 90 days from the date of receipt.

5.4 EXEMPTED LMO / GMO USE ACTIVITIES

a. Principal investigator (PI) for research involving exempted LMO / GMO from notification as in the First Schedule (Regulation 2) of the Biosafety *(Approval and Notification) Regulations 2010* must fill out the **UTHM / OSHE / UBK.001 REV 4** form (APPENDIX B) and submit to the Biosafety Unit OSHE.
b. The form submitted will be reviewed by the Biosafety Unit OSHE and brought to the IBC Chairman for approval.
c. Researchers must provide additional information regarding research if requested by the IBC Chairman.
d. The IBC Chairman will issue the decision on the application within 60 days from the date of receipt.

5.5 IBC ASSESSMENT

a. Laboratories visit for IBC Assessment will be conducted by BSO and IBC members.
b. Based on the assessment, researchers must take corrective action and submit relevant reports
c. IBC Assessment Report by BSO will use the **IBC / AP / 10 / ANNEX2** form provided by NBB.
d. Summarise the flow of approval that PIs need to follow when they are dealing with biological agents (LMO/GMO) as in **APPENDIX E**.

5.6 APPEALS

a. Researchers can appeal for any decision issued by the NBB or IBC.
b. Among the decisions that can be considered for appealing are: -
    i. Rejection of approval application.

ii. Imposition of terms or conditions.
iii. Imposition of additional terms or conditions.
iv. Postponement of approval.
v. Revocation of approval.
vi. Correction to approval.
vii. Rejection of application on factors that are not in line with the conditions of approval.
viii. Request for information, details or additional documents by NBB.
ix. Termination of activities.
x. Corrections to notifications.

c. An appeal against the decision of the NBB or IBC should be made by written letter/notice within 30 working days from the date the decision is issued.
d. The grounds of appeal and other relevant documents must be submitted within 30 working days after the notice of appeal is issued.

5.7 NOTICE OF EXTENSION AND TERMINATION OF RESEARCH

a. The flow chart for the Extension and Termination Process for Approved Research is as in **Appendix F**.
b. Notice to extend the term and terminate the research (Project Continuation & Termination Notification) must use the **IBC/PE-NT/10/ANNEX5** form provided by NBB and submitted to the Biosafety Unit OSHE for IBC and NBB approval.

5.8 PENALTIES

Any researcher involved in LMO / GMO research without obtaining the approval of notification via IBC and NBB will be subjected to legal actions, with legal penalties paid to the court of law.

a. Individual Penalty up to RM250,000 and/or imprisonment not exceeding five (5) years.
b. Corporate sector/Organisation Penalty (in this case, UTHM) does not exceed RM500,000.
c. If the offense is a continuing offense, the next fine will not exceed:
    i. RM 10,000 for individuals.
    ii. RM 20,000 for corporate/organisation

SECTION 6: CONTROLS FOR BIOHAZARDS IN LABORATORY ANIMALS

6.1 UTHM ANIMAL RESEARCH ETHICAL GUIDELINES & LAWS IN MALAYSIA

The use of living or dead animals in experiments and research or any related works must fully adhere to moral and ethical obligations as outlined in the animal ethics research guideline issued by the research management centre (Guidelines for animal and plant research ethics Universiti Tun Hussein Onn Malaysia 2018). The Principal Investigator and all personnel involved must be aware of the biosecurity threats posed when working with animals regardless of the researchers themselves, colleagues, students or community. In addition, the principal researcher must also comply with other laws governing animals in Malaysia from time to time such as:

a. Animal Act 1953

An act to amend and consolidate the laws for preventing the introduction into, and the spreading within, Peninsular Malaysia of diseases of animals; for the control of the movement of animals into, within and from Peninsular Malaysia; for the control of the slaughter of animals; for the prevention of cruelty to animals; for measures pertaining to the general welfare, conservation and improvement of animals in Peninsular Malaysia; and for purposes connected therewith.

b. Animal Welfare Act 2015

An act to provide for the establishment of the Animal Welfare Board, to set out the functions of the Board, to promote the welfare and responsible ownership of animals, and for related matters.

c. Feed Act 2009

An act to establish the Feed Board, to regulate feed quality by controlling the importation, manufacture, sale and use of feed and feed additive, to ensure that feed satisfies the nutritional requirement of animals, is not harmful to animals and is not contaminated so that animals and animal products are safe for human consumption and other usage, and for other matters incidental thereto.

6.2 RESPONSIBILITY OF PRINCIPAL INVESTIGATOR

The principal investigator (PI) is responsible for implementing the procedures designed to prevent biohazards' exposure to or transmission from laboratory animals to humans. PI must be aware of naturally occurring diseases of laboratory animals transmissible to humans and experimentally induced diseases, which may be harmful to humans. The primary responsibility for reducing or eliminating such risks lies with the PI.

Procedures designed for PI include handling and disposal of animals, contaminated animals, animal wastes and other related risks to protect the well-being of all personnel involved while at the same time maintaining the integrity of the experimental program and minimising risk to people participating in this program as well as animals. To achieve this, PI must be an expert in understanding the potential hazards involved in working with animals. All decisions made by PI regarding the selection of equipment, facilities and procedures are to minimise the risk involved in working with animals. A well-designed program that incorporates both animal care program and animal facility is compulsory to reduce biohazard exposure in animal facilities. This manual serves only as guidelines and is unable to outline all the definitive procedures and potential biohazard risks arising from working with animals and beyond the scope of this manual.

As outlined by UTHM animal research ethics guidelines, PI is responsible for notifying and providing the IBC and the Research Management Centre (RMC) of UTHM with specific information to their personnel on the biohazardous agents involved (carcinogen, radioactive isotope, etc.), the host range, the ability of experimentally infected animals to infect non-exposed animals or to excrete the agent in urine or faeces, special caging or animal isolation requirements, the need to autoclave isolation cages and their content prior to processing, and the selection and use of appropriate PPE

6.3 ANIMAL BIOSAFETY LEVEL

Animal research facility/laboratory used for animal research is best to be an independent and detached unit from other laboratories or working spaces. This is quite difficult to achieve in UTHM scenario and if it adjoins other area such as laboratory/working space, an isolation room/space should be available for the purpose of decontamination and disinfestation. Animal biosafety guideline introduced by WHO outlines four biosafety containment levels referred to as Animal Biosafety Lab (ABSL) (**Table 6.1**). However, only ABSL-1 and ABSL-2 are applicable at UTHM.

Table 6.1 . Animal facility containment levels: summary of practices and Safety equipment

Risk Group	Containment Level	Laboratory Practices and Safety Equipment
1	ABSL-1	Limited access, protective clothing and gloves
2	ABSL-2	ABSL-1 practices plus: hazard warning signs. Class I or II BSCs for activities that produce aerosols. Decontamination of waste and cages before washing
3	ABSL-3	ABSL-2 practices plus: controlled access. BSCs and special protective clothing for all activities
4	ABSL-4	ABSL-3 plus: strictly limited access. Clothing change before entering. Class III BSCs or positive pressure suits. Shower on exit. Decontamination of all wastes before removal from the facility

* ABSL, Animal Facility Biosafety Level; BSCs, Biological Safety Cabinets

The biosafety levels (facilities, practices, and operational requirements) recommended for working with biohazardous agents in vivo and in vitro are comparable. All of the facility requirements discussed for the biosafety laboratories in the previous sections are applicable to research with animals as well. Similar to BSL, ABSL considers different agents and animals to be used in the laboratory. In terms of agents, the factors include the normal route of transmission, the volumes and concentrations used, the route of inoculation, and the route the agents can be excreted. Regarding animals used in the animal

laboratory, factors to consider include the nature of the animals, i.e., their aggressiveness and tendency to bite and scratch, natural ectoparasites and endoparasites, and zoonotic diseases which they are susceptible and possible dissemination of allergens. Animals that have received a biohazardous agent should be housed in separate animal rooms, preferably in limited access rooms on a separate ventilation system. Animal room doors and individual cages should be conspicuously labelled with information regarding the agent used, the date of exposure, the biohazard symbol, and the names and telephone numbers of PI and responsible technician. As outlined by WHO animal biosafety guidelines, the followings are the criteria for the animal facility:

a. Animal Facility – Biosafety Level 1

ABSL-1 is necessary for the maintenance of most stock animals after quarantine (except for nonhuman primates, on which national authorities should be consulted) and for animals that are purposely inoculated with agents in Risk Group 1, in which good microbiological practice (GMP) are required. The ABSL coordinator must establish protocols, policies, and procedures for all operations, including access to the vivarium. A suitable medical surveillance programme for all personnel must be instituted with regard to the operation of this facility. Operation manual and safety manual must be carefully designed and strictly adopted by all personnel.

b. Animal facility – Biosafety Level 2

ABSL-2 is necessary for any work related to animals that are deliberately inoculated with microorganisms in Risk Group 2. In this situation, the following safety precautions apply:

i. All the requirements for ABSL-1 must be met.
ii. Biohazard warning signs should be posted on doors and other appropriate places.
iii. The facility must be designed for easy cleaning and housekeeping.
iv. Doors must open inwards and be self-closing.
v. Heating, ventilation and lighting must be adequate.
vi. If mechanical ventilation is provided, the airflow must be inwards. Exhaust air is discharged to the outside and should not be recirculated to any part of the building.
vii. Access must be restricted only to authorised persons.
viii. No animals should be admitted other than those for experimental use.

ix. There should be an arthropod and rodent control programme.
x. Windows, if present, must be secure, resistant to breakage and, if able to be opened, must be fitted with arthropod-proof screens.
xi. After use, work surfaces must be decontaminated with effective disinfectants
xii. Biological safety cabinets (Classes I or II) or isolator cages with dedicated air supplies and HEPA-filtered exhaust air must be provided for work that may involve the generation of aerosols.
xiii. An autoclave must be available on-site or in appropriate proximity to the animal facility.
xiv. Animal bedding materials must be removed in a manner that minimises the generation of aerosols and dust.
xv. All waste materials and bedding must be decontaminated before disposal.
xvi. The use of sharp instruments should be restricted whenever possible. Sharps should always be collected in puncture-proof/-resistant containers fitted with covers and treated as infectious.
xvii. Material for autoclaving or incineration must be transported safely, in closed containers.
xviii. Animal cages must be decontaminated after use.
xix. Animal carcasses should be incinerated.
xx. Protective clothing and equipment must be worn in the facility and removed on leaving.
xxi. Hand-washing facilities must be provided. Staff must wash their hands before leaving the animal facility.
xxii. All injuries, however minor, must be treated appropriately, reported and recorded.
xxiii. Eating, drinking, smoking and application of cosmetics must be forbidden in the facility.
xxiv. All personnel must receive appropriate training

6.4 ANIMAL BLOOD AND BLOOD PRODUCTS

This section describes how to work safely with non-human primate and animal (non-primate) blood, body fluids, tissues, and cell lines.

a. Non-Human Primate Blood, Body Fluids, Tissues and Cell Lines

Investigators working with non-human primates or non-human primate blood, body fluids, tissues, and cell lines should be concerned about safe handling because of the extreme severity of some

agents that primates can harbour without showing any clinical disease. The same blood and body fluid precautions used for humans must be consistently observed with all specimens from non-human primates. All laboratory personnel must be familiar with these precautions before working with primate body fluids.

b. Animal (non-primate) Blood, Body Fluids and Tissue, and Cell Lines

Non-primates generally present a less immediate hazard potential than primates. However, bats, dogs, cats, rabbits, rats, mice, etc. can carry microorganisms that are infectious to humans. In particular, animals acquired from unregulated sources must be considered a potential source of infection. For example, dogs and cats can carry rabies. Other infectious agents may be present without producing clinical illness in the animal. Generally, the same good laboratory practices used when working with primate source materials are followed when working with non-primate blood, body fluids, and tissue.

6.5 ANIMAL WORK PRACTICES AND ENGINEERING CONTROLS

The following work practices and engineering controls apply in addition to the biosafety practices discussed in the previous section of this manual.

a. **Gloves** – Personnel who handle animals must wear gloves appropriate for the task. Wash hands after removing gloves.
b. **Additional PPE** – Personnel handling animals that have received biohazardous agents must wear a face mask, gloves, and gown or other appropriate PPE.
c. **Animal cages** – Animals that are infected with a biohazardous agent are isolated within specific barriers such as filter-top cages, isolation racks, or ventilated racks. In all of these systems, the effectiveness of the barrier is determined by its design and the personnel using it. Thus, employee training is of paramount importance.
d. **Transport of animals** – Extreme care must be taken in transferring animals from the biohazard animal rooms to the laboratories or other facilities. The animal must be in a sealed container or filter-top cage, and transport equipment must be sanitised or sterilised immediately after transport.
e. **Necropsy** – Personnel conducting necropsies must wear appropriate PPE. Post-mortem examinations of small animals exposed to biohazards should be conducted in Class II BSCs when

possible. If such equipment is not available, extreme care must be taken to guard against the creation of aerosols and the contamination of conventional necropsy facilities. The necropsy table should be stainless steel and have suitable flushing devices. An appropriate disinfectant should be used to completely and thoroughly disinfect all instruments and working surfaces that come into contact with animal tissues.

f. **Perfusions** – Perfusions of animals infected with biohazardous agents must be performed in a fume hood or a non-recirculating BSC.

6.6 ANIMAL WASTE HANDLING PROCEDURES

Animal waste collection and disposal should be scheduled on a regular and timely basis. When storage of animal waste is required, the area selected should be physically separated from other storage facilities and free of insects and rodents. Refrigerated storage facilities are recommended when waste must be held for more than four to six hours.

a. **Disposal of Animal Carcasses and Body Parts**

Animal carcasses and animal body parts are a type of biological waste that requires special handling depending on whether it is radioactive, infectious, or non-hazardous. Consult Animal Research Waste Flow Chart (**Figure 6**) to determine how to dispose of animal carcasses and parts.

b. **Disposal of Animal Blood and Blood Products**

Animal blood, blood products and animal waste/bedding from animals infected with recDNA or other biohazardous agents are handled as biomedical waste that can be chemically decontaminated or autoclaved according to established guidelines prior to disposal. In particular, blood, blood products, tissue, and tissue suspension, including blood-contaminated items, must be decontaminated prior to disposal. The exempted are small amounts of non-primate blood, which can be flushed down sink drains without chemical treatment.

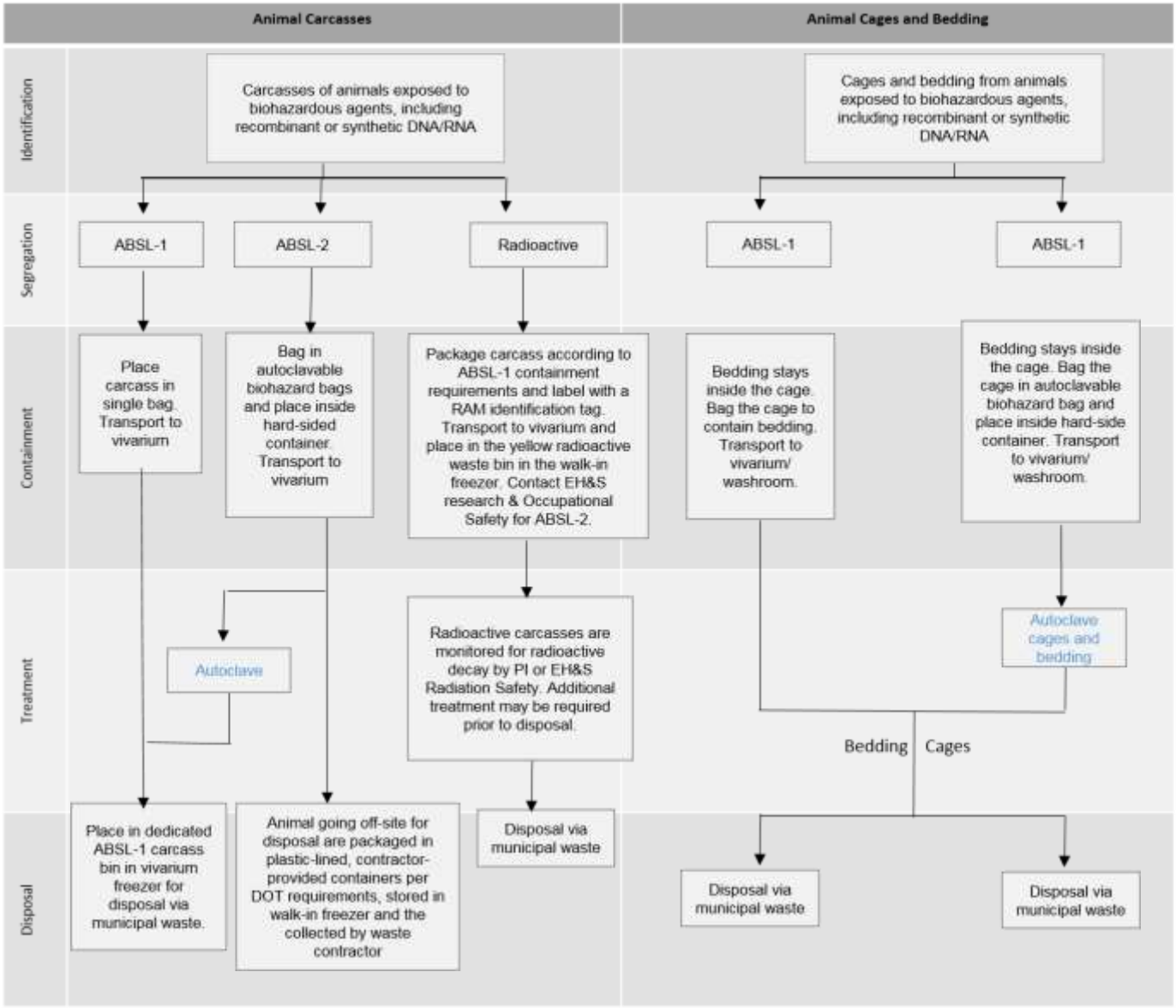


Figure 6.1. Animal Research Waste Flow Chart

BORANG LAPORAN KEMALANGAN, KEJADIAN BERBAHAYA, PENYAKIT PEKERJAAN DAN KERACUNAN PEKERJAAN

1. BUTIRAN KEJADIAN

i. Tarikh :_____

ii. Masa :_____

iii. Lokasi :_____

iv. PTj :_____

v. Jenis Kejadian :

☐ Kemalangan

☐ Kejadian Berbahaya

☐ Penyakit Pekerjaan

☐ Keracunan Pekerjaan

2. BUTIRAN MANGSA *(Sila gunakan lampiran sekiranya mangsa lebih dari seorang)*

i. Nama :_____ ii. Umur :_____

iii. No. K.P/ No.Paspot :_____ iv. Jantina :_____

v. Status : ☐ Staf ☐ Pelajar ☐ Pekerja Kontrak ☐ Orang Awam

3. KECEDERAAN DAN RAWATAN

i. Nama Klinik/Hospital :_____

ii. Jenis Rawatan Yang Diberikan :_____

iii.Bahagian Anggota Yang Tercedera :_____

iv. Cuti Sakit :_____ hari (Tarikh _____)

4. PERINCIAN KEJADIAN

(Sila jelaskan secara terperinci keadaan sebelum, semasa dan selepas Kemalangan/ Kejadian Berbahaya/ Penyakit Pekerjaan/ Keracunan Pekerjaan ini berlaku. Gunakan lampiran sekiranya ruangan yang disediakan tidak mencukupi.)

5. KEROSAKAN HARTA BENDA

(Sila nyatakan harta benda dan anggaran kos yang terlibat)

6. CADANGAN PENAMBAHBAIKAN

(Sila nyatakan cadangan penambahbaikan yang diambil bagi mengelakkan kejadian daripada berulang.)

7. PERAKUAN LAPORAN	
Saya mengaku segala butir-butir dan maklumat yang saya berikan di atas adalah benar.	
Disediakan oleh :	Disahkan oleh :
_____	_____
(Pelapor)	(SLO/ Setiausaha JKKP PTj)
Nama : No. K.P : Tarikh :	Nama : Tarikh :
**Sila lampirkan dokumen sokongan seperti sijil cuti sakit dan/ atau bukti bergambar berkaitan dengan kejadian.*	

8. ULASAN DAN PENGESAHAN KETUA PTj
Ulasan Ketua PTj :
_____ Tarikh : _____
(Tandatangan & Cop Rasmi Ketua PTj)

Perhatian:

Sila majukan satu salinan borang ini yang telah lengkap diisi kepada Bahagian Persekitaran, Keselamatan dan Kesihatan Pekerjaan (OSHE) untuk tujuan penyimpanan rekod.

PEMBERITAHUAN AKTIVITI KEGUNAAN TERKAWAL ORGANISMA HIDUP (LO) DAN ORGANISMA HIDUP YANG DIUBAH SUAI (LMO) DI UNIVERSITI

NOTIFICATION FOR CONTAINED USE ACTIVITY OF LIVING ORGANISM (LO) AND LIVING MODIFIED ORGANISM (LMO) IN THE UNIVERSITY

A. Maklumat Am *Preliminary Information*		
Nama Pejabat / Fakulti / PTj *Office / Faculty / PTj Name*	:	
Nama Pemohon (Penyelidik Utama) *Name of applicant (Principal Investigator)*	:	
Jawatan *Position*	:	
Nombor Telefon *Telephone no.*	:	
E-mel *E-mail*	:	

B. Maklumat LO/LMO *LO/LMO Information*		
LO/LMO	**Tahap Pembendungan** *Containment Level* *(Level 1 , 2 , 3, 4)*	**Kumpulan Risiko** *Risk Group* *(RG 1, 2, 3, 4)*

C. Pengurusan Risiko /*Risk Management*	
Bagi penyediaan SOP, Sila gunakan Templat SOP yang disediakan. *Use the template of SOP provided.* (UTHM/OSHE/UBK.006)	
1.	Adakah anda bercadang untuk mengangkut LO / LMO ke luar dari premis atau di antara premis? Jika ya, berikan Prosedur Operasi Standard (SOP) tertentu yang mematuhi Garis Panduan Biokeselamatan. *Do you propose to transport the LO/LMO outside the premises or between premises? If yes, provide specific Standard Operating Procedures (SOPs) which are in compliance with Biosafety Guidelines. Use the template of SOP provided*
2.	Bagaimana LO / LMO akan dilupuskan? *How will the LO/ LMO be disposed of?*

3.	Bagaimanakah sisa pepejal dan cecair daripada aktiviti tersebut dirawat dan dilupuskan? (contoh: media, objek tajam, sarung tangan pakai buang, dan lain-lain). Berikan Prosedur Operasi Standard (SOP) tertentu yang mematuhi Garis Panduan Biokeselamatan. *How will the solid and liquid wastes from the activities be treated and disposed of? (e.g. media, sharps, disposable gloves, etc.) Provide specific Standard Operating Procedures (SOPs) which are in compliance with Biosafety Guidelines.*
4.	Bagaimanakah kaedah / lokasi penyimpanan LO / LMO tersebut? *What is the method / location of the LO / LMO storage?*

D. Pelan Tindakan Kecemasan (*Emergency Response Plan)*
Sertakan Prosedur Operasi Standard (SOP) tertentu yang mematuhi Garis Panduan Biokeselamatan sekiranya berlaku tumpahan atau pelepasan yang tidak disengajakan bagi LO/LMO tersebut. *Provide specific Standard Operating Procedures (SOPs which are in compliance with Biosafety Guidelines in the case of an unintentional release or accidental spill of the LO/LMO (e.g.to contain and treat spillage.)*

E. Pengakuan *(Declaration)*	
Kami mengakui bahawa semua maklumat dan dokumen diberikan di sini adalah benar. Kami memahami bahawa memberikan maklumat yang mengelirukan kepada Jawatankuasa Keinstitusian Biokeselamatan (IBC), dengan sengaja atau sebaliknya, adalah suatu kesalahan di bawah Akta Biokeselamatan 2007. *We declare that all information and document herein are true and correct. We understand that providing misleading information to the Institutional Biosafety Committee (IBC), deliberately or otherwise, is an offence under the Biosafety Act 2007.*	
i. Pemohon/ Penyelidik Utama : *Applicant / Principal Investigator* _____ Nama (*Name*) : Tarikh *Date*) : Cop Rasmi (*Official stamp)*:	**ii. Pegawai Biokeselamatan**: *Biosafety Officer* _____ Nama (*Name*) : Tarikh *Date*) : Cop Rasmi (*Official stamp)*:

APPENDIX C
UTHM/OSHE/UBK.006

STANDARD OPERATING PROCEDURE
JAWATANKUASA BIOKESELAMATAN UNIVERSITI (IBC)

Project Tittle :	
P.I.C :	
Faculty :	

(NAME OF THIS STANDARD OPERATING PROCEDURE)

Standard Operating Procedure No.	
Revision No:	
Original Date of Issue:	
Revision Date:	
Revised by:	
Approved by:	

Background: (What requirements will this standard operating procedure meet?)

Purpose: To provide instruction on …

Related Standards and Procedures:

- List any related standards, good operating practices or other standard operating procedures.
-

Procedure:

- List the tasks step by step to provide instruction on how to perform this procedure.
-

-

Related Forms and documentation:

- List the forms pertaining to this procedure.
-

Records:

- List the records that will be kept as a result of this procedure.

APPENDIX D

UTHM/OSHE/UBK.005

LABORATORY BIOSAFETY CHECKLIST

FOR BIOSAFETY LEVEL 1 AND 2

A. Premise Information	
1.0	Premise :
2.0	Principal Investigator :
3.0	Inspector :
4.0	Date / Time :
5.0	Laboratory Biosafety Level: **1 / 2**

Check (√) in the appropriate box that most accurately describes the laboratory in which the work will be performed.

B. Biosafety Level 1					
1.0	**Laboratory Facilities**	***Yes***	***No***	***NA***	***Details/ Comment***
1.1	Is a universal biohazard symbol for BSL 1 posted at the laboratory entrance? Does the sign include the name and phone number of the laboratory supervisor or other responsible personnel?				
1.2	Do the laboratories have a sink for hand washing?				
1.3	Are furniture in the laboratory capable of supporting anticipated loads and uses?				
1.4	Are spaces between benches, cabinets and equipment accessible for cleaning?				
1.5	Are bench tops impervious to water and resistant to heat, organic solvents, acids, alkalis, and other chemicals?				

1.6	Are furniture including bench tops in the laboratory covered with a non-porous material can be easily cleaned?				
1.7	Are laboratory windows that are open to the exterior fitted with screens?				
1.8	Is an autoclave for pre-treatment of laboratory wastes available?				
1.9	Is an eyewash fountain readily available in the laboratory?				
1.10	Is an effective integrated pest management programme in place and managed appropriately?				
2.0	**Safety Equipment**	**Yes**	**No**	**NA**	**Details/ Comment**
2.1	Is there a first aid kit available?				
2.2	Are suitable PPE available and used by laboratory personnel?				
2.3	Is there any storage equipment to keep GM materials? (Example: refrigerator)				
2.4	Is there a fume hood for working with hazardous chemicals?				
2.5	Are the equipment regularly maintained?				
3.0	**Work Practices**	**Yes**	**No**	**NA**	**Details/ Comment**
3.1	Is there any biohazardous materials handled in the laboratory?				

3.2	Do personnel wash their hands before leaving the lab?				
3.3	Are mechanical pipetting devices used?				
3.4	Is there signage available prohibiting eating, drinking, smoking, handling contact lenses, applying cosmetics, and storing food for human consumption in the lab?				
3.5	Are work surfaces decontaminated with an effective disinfectant on completion of work, especially after spills or splashes of biohazardous materials?				
3.6	Is a sharp waste bin available for disposal of all syringes/ needles/ sharp items?				
3.7	Are re-usable sharp items properly cleaned and disinfected?				
3.8	Is there a biological spill kit available?				
3.9	Are all wastes that are contaminated with biohazardous materials autoclaved or decontaminated?				
3.10	Have all personnel been provided information about hazards and risks about their work activity?				
3.11	Are laboratory waste segregated into dedicated waste bins and labelled properly?				
4.0	**Records and Documents**	**Yes**	**No**	**NA**	**Details/ Comment**
4.1	Procurement and transfers of GMO/LMO				

4.2	Biological material /LMO inventory				
4.3	SOPs for contained use activity				
4.4	Staff training and competency				
4.5	Equipment maintenance				
4.6	Decontamination and validation				

C. Biosafety Level 2					
1.0	**Laboratory Facilities**	***Yes***	***No***	***NA***	***Details/ Comment***
1.1	Is a universal biohazard symbol for BSL 2 posted at the laboratory entrance? Does the sign include the name and phone number of the laboratory supervisor or other responsible personnel?				
1.2	The design of the facility should be made as such that laboratory activities are separated from the common areas. (Examples: offices and pantry)				
1.3	Is a designated hand basin of the hands-free operation type provided in each laboratory?				
1.4	Are furniture in the laboratory capable of supporting anticipated loads and uses?				
1.5	Are spaces between benches, cabinets and equipment accessible for cleaning?				

1.6	Are the benchtops impervious to water and made resistant to heat, organic solvents, acids, alkalis, and other chemicals?				
1.7	Are furniture including benchtops in the laboratory covered with a non-porous material that can be easily cleaned?				
1.8	If the windows are present, are they always closed?				
1.9	Is an autoclave for pre-treatment of laboratory wastes available in the contained facility?				
1.10	Is an eyewash fountain readily available in the laboratory?				
1.11	Is an effective integrated pest management programme in place and managed appropriately?				
1.12	Are laboratory floors smooth, easy to clean and resistant to chemicals?				
1.13	Does the ventilation in the laboratory have directional air flow into the laboratory areas?				
1.14	Is there good housekeeping in the laboratory?				
2.0	**Safety Equipment**	**Yes**	**No**	**NA**	**Details/ Comment**
2.1	Is there a first aid kit available?				
2.2	Is there a Class II biological safety cabinet in the laboratory, certified annually?				
2.3	Is the BSC suitably located, away from the door and air vent?				

2.4	Is the BSC free of equipment or supplies that can block the air grills and disrupt proper airflow?				
2.5	When lab personnel use vacuum lines with biohazardous materials, are they protected with High Efficiency Particulate Air (HEPA) filters?				
2.6	Is equipment for use or storage of biohazardous materials (i.e. refrigerator, freezers) labelled with a biohazard symbol?				
2.7	Are GM/LMOs kept separately from non-GM/LMOs?				
2.8	Is there a fume hood for working with hazardous chemicals?				
3.0	**PPE (Personal Protective Equipment)**	**Yes**	**No**	**NA**	**Details/ Comment**
3.1	Are suitable PPE available for laboratory hazards and used by laboratory personnel?				
3.2	Are respiratory masks available for infectious aerosol work?				
3.3	Are closed-toe shoes used in the laboratory?				
4.0	**Work Practices**	**Yes**	**No**	**NA**	**Details/ Comment**
4.1	Is there any biohazardous material handled in the laboratory?				
4.2	Do personnel wash their hands before leaving the laboratory?				
4.3	Are mechanical pipetting devices used?				

4.4	Is signage available to prohibit eating, drinking, smoking, handling contact lenses, applying cosmetics, and storing food for human consumption in the laboratory?				
4.5	Are work surfaces decontaminated with effective disinfectant after completion of work especially after spills or splashes of biohazardous materials?				
4.6	Is there a sharp bin available for disposal of and all syringes/ needles/ sharps?				
4.7	Are re-usable sharps properly cleaned and disinfected?				
4.8	Is there a biological spill kit available?				
4.9	Are all wastes that are contaminated with biohazardous materials autoclaved or decontaminated?				
4.10	Have all personnel been provided information about hazards and risks of their work activities?				
4.11	Is laboratory waste segregated into dedicated waste bins and properly labelled?				
4.12	Is a suitable chemical disinfectant used to inactivate liquid waste?				
4.13	Is a medical surveillance program available for laboratory personnel? (Example: Hep B vaccination)				

4.14	Is there a Laboratory Biosafety Guidelines or Biosafety Manual available in the laboratory?				
4.15	Are the lab-specific Biosafety procedures included in the Laboratory Biosafety Guidelines or Biosafety Manual?				
4.16	Are needle-locking syringes or safety hypodermic needles used when appropriate?				
4.17	Are biohazardous materials transported in covered containers to prevent leakage?				
4.18	Is there any incident/accident/laboratory exposure reporting system available?				
4.19	Is there an ERP available?				
4.20	Is medical follow-up obtained appropriate?				
4.21	Are animals and plants unrelated to the experiment prohibited from the laboratory?				
4.22	Are experiments involving animals and plants carried out in the laboratory?				
5.0	**Records and Documents**	**Yes**	**No**	**NA**	**Details/ Comment**
5.1	Procurement and transfers of GMO/LMO				
5.2	Biological material /LMO inventory				
5.3	SOPs for contained use activity				
5.4	Staff training and competency				

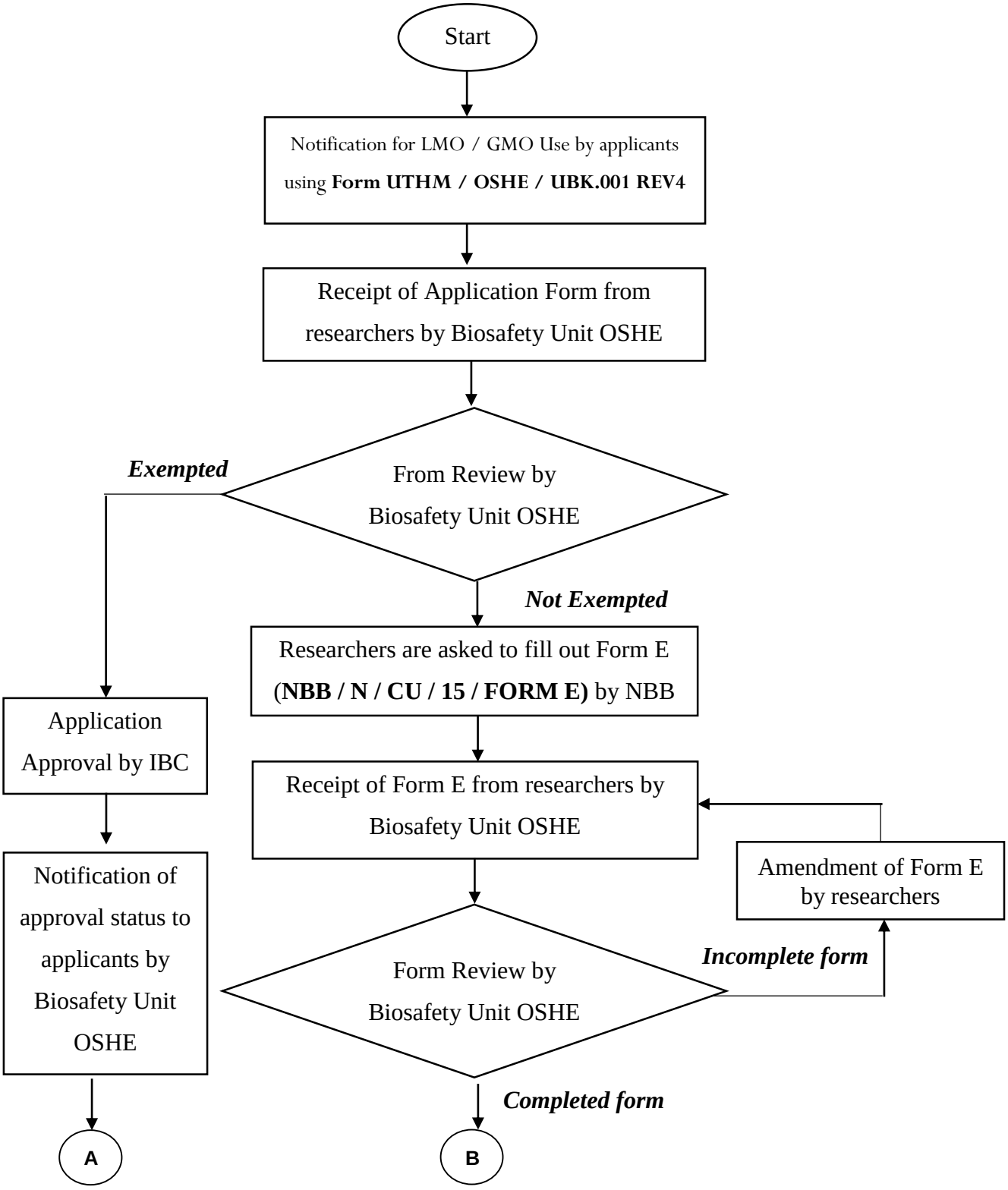
5.5	Equipment maintenance				
5.6	Decontamination and validation				
5.7	Incident/accident/laboratory exposure				

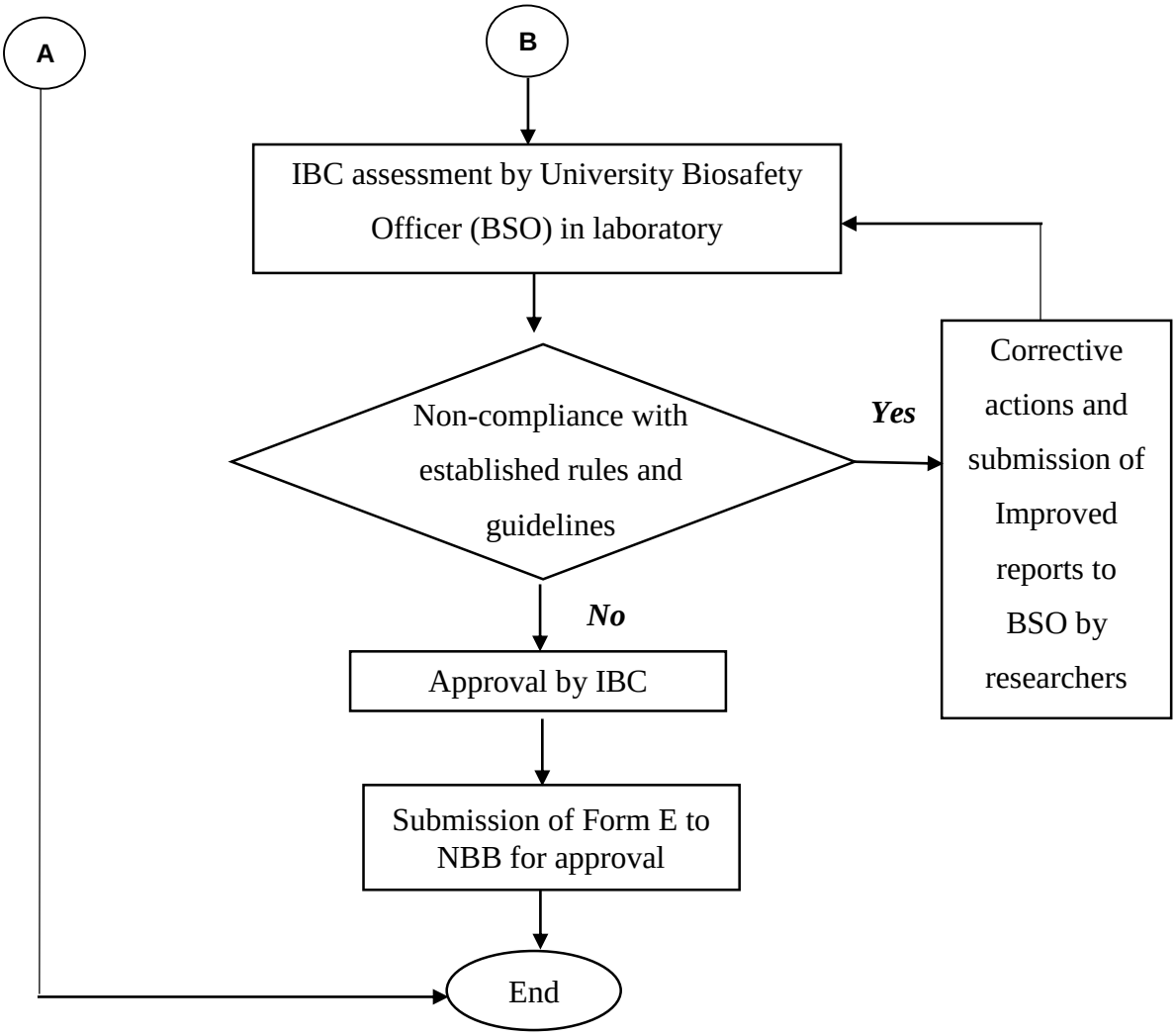
D. Additional comments/observations and recommendation

Inspector’s Signature:

Date:

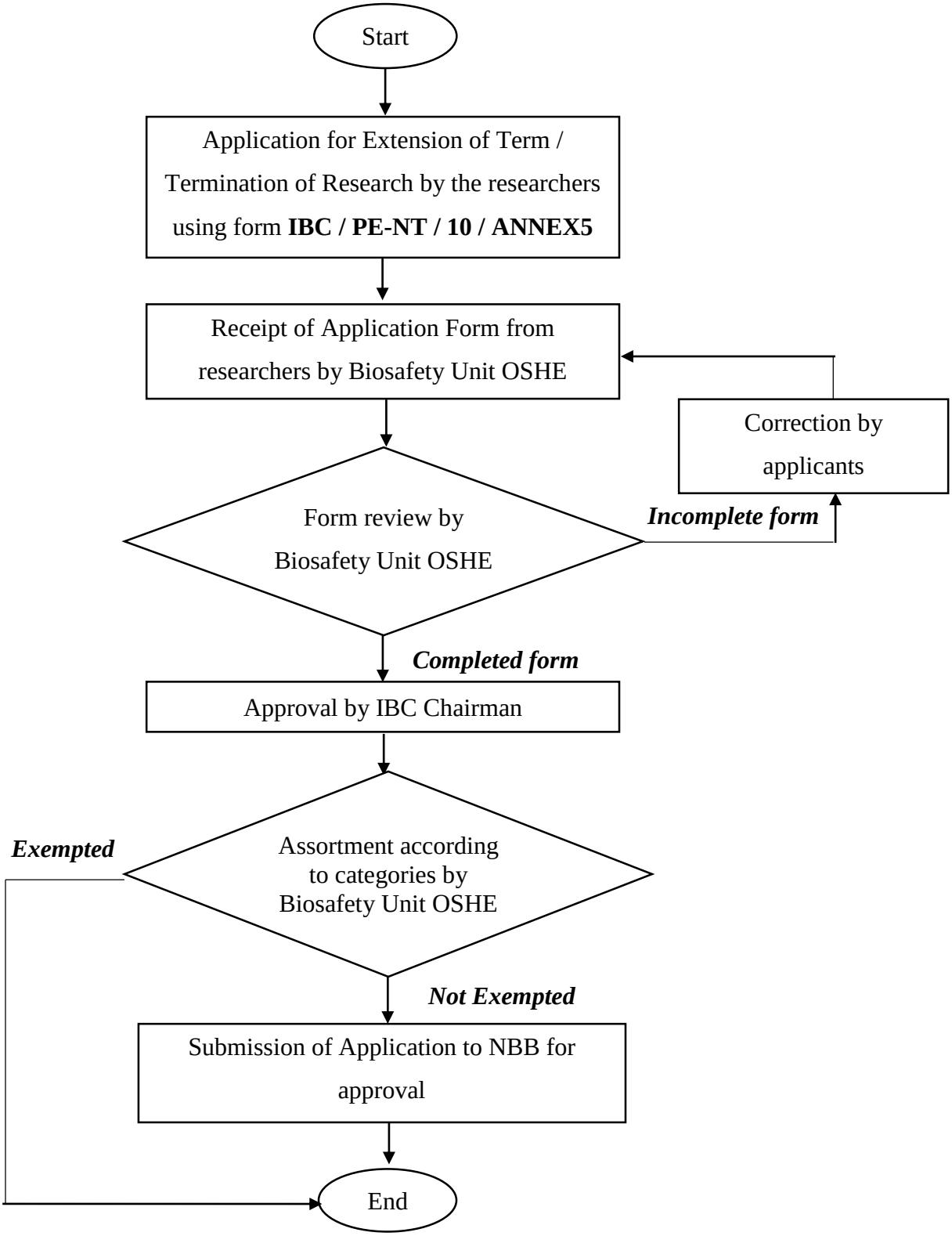
Flowchart of Notification of LMO / GMO Use in Research



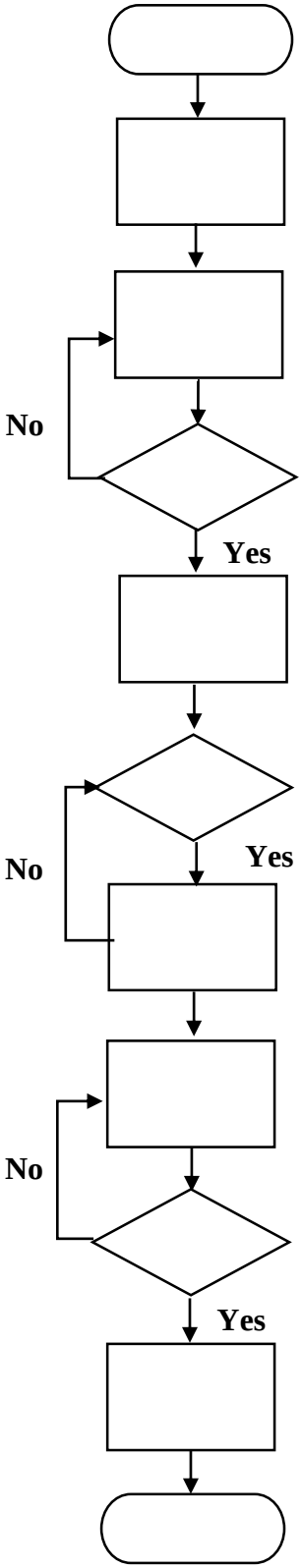

A
B
IBC assessment by University Biosafety Officer (BSO) in laboratory
Non-compliance with established rules and guidelines
Yes
Corrective actions and submission of Improved reports to BSO by researchers
No
Approval by IBC
Submission of Form E to NBB for approval
End

APPENDIX F

Flow Chart of Research Extension and Research Termination



Flow Chart of Purchasing Microorganisms



START

Application for acquisition of living organism and microorganism by the applicant to PTj.

Application for acquisition of living organism and microorganism by the applicant to OSHE.

Fill out the registration form (notification) as found in Appendix B. Approved.

Delivery of living organisms and microorganisms by supplier.

Living organisms and microorganisms shipped by suppliers meet legal requirements. Passed.

Applicant ensures living organisms and microorganisms are stored according to proper storage methods.

Submission of procurement payment form by the applicant to PTj together with a checklist of legal requirements.

Payment process to supplier. Passed.

Complete the notification form and the relevant SOP to OSHE.

END

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