TARLETON STATE UNIVERSITY Biological Safety Program

Program Name: Biological Safety
Department Name: TSU Risk Management & Compliance

Doc. No.: BIOS-04-L2-S0-CH0-001 Rev. No.: 3

	Tarleton State University			
		Program:	Biological Safety	
	Risk Management & Compliance Level 2	Doc. No.:	BIOS-04-L2-S0-CH0-001	
		Rev No:	3	
		Date:	8-31-2018	
	Level 2	Dept.:	TSU Risk Management & Compliance	

Concurrence and Approval

This Environmental Management System Document was developed for use by all Tarleton State University Employees and has been reviewed and approved by the following approvers.

Document Custodian:

Hector C. Davis, Environmental Health and Safety Coordinator

	Tarleton State University				
		Program:	Biological Safety		
	Risk Management & Compliance	Doc. No.:	BIOS-04-L2-S0-CH0-001		
		Rev No:	3		
		Date:	8-31-2018		
	Level 2	Dept.:	TSU Risk Management & Compliance		

Change History

Revision	Interim	Effective Date	
Number	Change No.		Description of Change
001	0	10-March-2014	Initial document release under new document and record control guidance
002	0	06-October-2016	Biannual review
003	0	31-August-2018	Biannual review

	Tarleton State University				
		Program:	Biological Safety		
	Risk Management & Compliance Level 2	Doc. No.:	BIOS-04-L2-S0-CH0-001		
		Rev No:	3		
		Date:	8-31-2018		
		Dept.:	TSU Risk Management & Compliance		

1. GENERAL

The following information is provided to assist Tarleton Departments in developing procedures to meet biological safety requirements to protect students, employees, and the environment.

Any pregnant students, or students planning to become pregnant, should consult their health care provider to determine what, if any, additional precautions are needed based on their individual situation. It is the responsibility of the student to communicate their needs to their immediate supervisor as soon as possible in order for risk-reduction to begin when it can be most effective, and to determine if additional modifications are necessary. While the university cannot mandate that the student notify it that she is pregnant or is planning to become pregnant, the university strongly recommends that students do provide notification so appropriate steps can be taken to ensure the health of both parent and child. To communicate health circumstances or to request additional information, please contact Tarleton's Title IX Coordinator within the Department of Employee Services at x9128.

2. PURPOSE

This program sets forth recommended minimum biological safety requirements that need to be followed to maximize the safety of all workers.

3. SCOPE

The Biological Safety Program information, guidelines and procedures are applicable to all Tarleton State University employees who work with biological materials.

4. CONTAINMENT

The primary principle of biological safety (i.e., biosafety) is containment. The term containment refers to a series of safe methods for managing infectious agents in the laboratory. The purpose of containment is to reduce or eliminate human and environmental exposure to potentially harmful agents.

a. Primary and Secondary Containment

There are two levels of biological containment--primary and secondary.

	Tarleton State University			
		Program:	Biological Safety	
	Risk Management & Compliance Level 2	Doc. No.:	BIOS-04-L2-S0-CH0-001	
		Rev No:	3	
		Date:	8-31-2018	
		Dept.:	TSU Risk Management & Compliance	

- i. Primary containment protects people and the immediate laboratory environment from exposure to infectious agents. Good microbial techniques and safety equipment provide sufficient primary containment. Examples of primary barriers include safety equipment such as biological safety cabinets, enclosed containers, and safety centrifuge cups. Occasionally, when it is impractical to work in biological safety cabinets, personal protective equipment, such as lab coats and gloves may act as the primary barrier between personnel and infectious materials.
- ii. Secondary containment protects the environment external to the laboratory from exposure to infectious materials. Good facility design and operational practices provide secondary containment. Examples of secondary barriers include work areas that are separate from public areas, decontamination facilities, handwashing facilities, special ventilation systems, and airlocks.

b. Elements of Containment

Ultimately, the three key elements of biological containment are laboratory practices, safety equipment, and facility design. To ensure minimal exposure, employees must assess the hazards associated with their work and determine how to apply the biosafety principles appropriately.

IMPORTANT: Employees working with infectious agents or potentially infectious materials must review the Bloodborne Pathogen Program and be aware of hazards associated with their work. These workers must be trained and proficient in biosafety procedures and techniques.

5. GENERAL BIOSAFETY GUIDELINES

Biohazardous materials require special safety precautions and procedures. Follow these guidelines when working with infectious agents:

a. Personal Hygiene Guidelines:

Wash your hands thoroughly, as indicated below:

- i. After working with any biohazard
- ii. After removing gloves, laboratory coat, and other contaminated protective clothing
- iii. Before eating, drinking, smoking, or applying cosmetics
- iv. Before leaving the laboratory area
- v. Do not touch your face when handling biological material.
- vi. Never eat, drink, smoke, or apply cosmetics in the work area.

	Tarleton State University			
		Program:	Biological Safety	
	Risk Management & Compliance	Doc. No.:	BIOS-04-L2-S0-CH0-001	
		Rev No:	3	
		Date:	8-31-2018	
	Level 2	Dept.:	TSU Risk Management & Compliance	

b. Clothing Guidelines:

- i. Always wear a wrap-around gown or scrub suit, gloves, and a surgical mask when working with infectious agents or infected animals.
- ii. Wear gloves over gown cuffs.
- iii. Never wear contact lenses around infectious agents.
- iv. Do not wear potentially contaminated clothing outside the laboratory area.
- v. To remove contaminated clothing, follow these steps:
- vi. Remove booties from the back.
- vii. Remove head covering from the peak.
- viii. Untie gown while wearing gloves.
 - ix. Remove gloves by peeling them from the inside out.
 - x. Remove the gown by slipping your finger under the sleeve cuff of the gown.

c. Handling Guidelines:

- i. Use mechanical piping devices.
- ii. Minimize aerosol production.
- iii. Add disinfectant to water baths for infectious substances.
- iv. Use trunnion cups with screw caps for centrifuging procedures. Inspect the tubes before use.
- v. Use secondary leak-proof containers when transporting samples, cultures, inoculated petri dishes, and other containers of biohazardous materials.

d. Syringes:

Avoid using syringes and needles whenever possible. If a syringe is necessary, minimize your chances of exposure by following these guidelines:

- i. Use a needle-locking or disposable needle unit.
- ii. Take care not to stick yourself with a used needle.
- iii. Place used syringes into a pan of disinfectant without removing the needles.
- iv. Do not place used syringes in pan containing pipets or other glassware that requires sorting.
- v. Do not recap used needles.
- vi. Dispose of needles in an approved sharp container.

e. Work Area:

- i. Keep laboratory doors shut when experiments are in progress.
- ii. Limit access to laboratory areas when experiments involve biohazardous agents.
- iii. Ensure that warning signs are posted on laboratory doors. These signs should include the universal biohazard symbol and the approved biosafety level for the laboratory.
- iv. Ensure that vacuum lines have a suitable filter trap.

	Tarleton State University				
		Program:	Biological Safety		
	Risk Management & Compliance Level 2	Doc. No.:	BIOS-04-L2-S0-CH0-001		
		Rev No:	3		
		Date:	8-31-2018		
		Dept.:	TSU Risk Management & Compliance		

- v. Decontaminate work surfaces daily and after each spill.
- vi. Decontaminate all potentially contaminated equipment.
- vii. Transport contaminated materials in leak-proof containers.
- viii. Keep miscellaneous material (i.e., books, handbags, etc.) away from contaminated areas.
 - ix. Completely decontaminate equipment before having maintenance or repair work done.

f. Universal Precautions:

Clinical and diagnostic laboratories often handle specimens without full knowledge of the material's diagnosis; these specimens may contain infectious agents. To minimize exposure, observe universal precautions when handling any biological specimen. Consider all specimens to be infectious and treat these materials as potentially hazardous.

6. CDC and NIH Biosafety Levels

The Centers for Disease Control (CDC) and the National Institute of Health (NIH) have established four biosafety levels consisting of recommended laboratory practices, safety equipment, and facilities for various types of infectious agents. Each biosafety level accounts for the following risk criteria: 1) Nature of work being conducted, 2) Transmissibility, 3) Infectivity, and 4) Severity of disease. Refer to Table 1.

- a. Biosafety Level 1 (BSL-1): facilities that work with defined and characterized strains of viable organisms that do not cause disease in healthy adult humans (e.g., Bacillus subtilis and Naegleria gruberi). Level 1 precautions rely on standard microbial practices without special primary or secondary barriers. Biosafety Level 1 criteria are suitable for undergraduate and secondary education laboratories.
- b. Biosafety Level 2 (BSL-2): facilities that work with a broad range of indigenous moderate-risk agents known to cause human disease (e.g., Hepatitis B virus, salmonellae, and Toxoplasma spp.). Level 2 precautions are necessary when working with human blood, body fluids, or tissues where the presence of an infectious agent is unknown. The primary hazards associated with level 2 agents are injection and ingestion. Most Tarleton State University research laboratories should comply with Biosafety Level 2 criteria.
- c. Biosafety Level 3 (BSL-3) facilities that work with indigenous or exotic agents with the potential for aerosol transmission and lethal infection (e.g.,

	Tarleton State University				
		Program:	Biological Safety		
	Risk Management & Compliance Level 2	Doc. No.:	BIOS-04-L2-S0-CH0-001		
		Rev No:	3		
		Date:	8-31-2018		
		Dept.:	TSU Risk Management & Compliance		

Mycobacterium tuberculosis). The primary hazards associated with level three agents are autoinoculation, ingestion, and inhalation. Level 3 precautions emphasize primary and secondary barriers. For primary protection, all laboratory manipulations should be performed in a biological safety cabinet or other enclosed equipment. Secondary protection should include controlled access to the laboratory and a specialized ventilation system.

d. Biosafety Level 4 (BSL-4) facilities that work with dangerous and exotic agents with a high risk of causing life-threatening disease, the possibility of aerosol transmission, and no known vaccine or therapy (e.g., Marburg or Congo-Crimean viruses). Level 4 agents require complete isolation. Class III biological safety cabinets or full-body air-supplied positive-pressure safety suits are necessary when working with level 4 agents. In addition, isolated facilities, specialized ventilation, and waste management systems are required. There are no Biosafety Level 4 facilities at Tarleton State University.

	Tarleton State University				
		Program:	Biological Safety		
4	Risk Management & Compliance Level 2	Doc. No.:	BIOS-04-L2-S0-CH0-001		
		Rev No:	3		
		Date:	8-31-2018		
		Dept.:	TSU Risk Management & Compliance		

Table 1: Summary of Laboratory Biosafety Levels for Infectious Agents

1 abie	1: Summary of Laborat	ory Biosafety Levels for			
BSL	Agents	Practices	Primary Barriers	Secondary Barriers	
1	Not known to cause disease in healthy adults.	Standard Microbiological Practices	None Required	Laboratory bench and sink required	
2	Agents associated with human disease • Routes of transmission include: percutaneous injury, ingestion, mucous membrane exposure	BSL-1 practices plus: Limited access Biohazard warning signs "Sharps" precautions Biosafety manual Defining any needed waste decontamination or medical surveillance policies	Primary Barriers: Class I or II BSCs or other physical containment devices used for all manipulations of agents that cause splashes or aerosols of infectious materials PPEs: Laboratory coats; gloves; face protection as needed	BSL-1 plus: Autoclave available	
3	Indigenous or exotic agents with the potential for aerosol transmission • Disease may have serious or lethal consequences	BSL-2 practices plus: Controlled access Decontamination of all waste Decontamination of laboratory clothing before laundering • Baseline serum collected and stored	Primary Barriers: Class I or II BSCs or other physical containment devices used for all open manipulation of agents PPEs: Protective laboratory clothing; gloves; respiratory protection as needed	BSL-2 plus: Physical separation from access corridors Self-closing, double door access Exhausted air not recirculated Negative airflow into laboratory	
4	Dangerous/exotic agents which pose high risk of life-threatening disease • Aerosol transmitted infections have occurred; or.related agents with unknown risk of transmission	BSL-3 practices plus: Clothing change before entering Shower upon exit All material decontaminated upon exit from facility	Primary barriers: • All procedures conducted in Class III BSCs or in Class I or II BSCs in combination with with full-body, air supplied, positive pressure personnel suit	BSL-3 plus: Separate building or isolated zone Dedicated supply/exhaust, vacuum, and decontamination systems Other requirements outlined in the text	

	Tarleton State University				
		Program:	Biological Safety		
	Risk Management & Compliance Level 2	Doc. No.:	BIOS-04-L2-S0-CH0-001		
		Rev No:	3		
		Date:	8-31-2018		
		Dept.:	TSU Risk Management & Compliance		

e. Animal Biosafety

Four biosafety levels are also described for infectious disease work with laboratory animals. Safety practices, equipment, and facilities are designated by Animal Biosafety Levels (ABSL) 1, 2, 3, and 4. Refer to Table 2.

Table 2: Summary of Laboratory Animal Biosafety Levels for Animal Experiments

ABSL	Agents	Practices	Primary Barriers	Secondary Barriers
1	Not known to cause	Standard animal care and	As required for normal care	Standard animal facility:
	disease in healthy	management practices,	of each species	No recirculation of exhaust
	adults.	including appropriate medical		air
		surveillance programs		Directional air flow
				recommended
				Hand washing sink
2	Agents associated	ADCI 1	ABSL-1 equipment plus	available ABSL-1 plus:
2	Agents associated with human disease	ABSL-1 practices plus: Limited access	primary barriers:	Autoclave available
	Hazard:	Biohazard warning signs	Containment equipment	Hand washing sink
	percutaneous injury,	"Sharps" precautions	appropriate for animal	available
	ingestion, mucous	Biosafety manual	species	Mechanical cage washer
	membrane exposure	• Decontamination of all	PPEs:	recommended
	•	infectious wastes and all	 Laboratory coats; gloves; 	
		animal cages prior to washing	face and respiratory	
			protection as needed	
3	Indigenous or exotic	ABSL-2 practices plus:	ABSL-2 equipment plus:	ABSL-2 plus:
	agents with the	Controlled access	Containment equipment for	Physical separation from
	potential for aerosol	Cages decontaminated before	housing animals and cage	access corridors
	transmission • Disease may have	bedding removed Decontamination of laboratory	dumping activities Class I, II or III BSCs	Self-closing, double door
	serious health effects	clothing before laundering	available for manipulative	Sealed penetrations
	serious neurin errects	• Disinfectant footbath as	procedures (inoculation,	Sealed windows
		needed	necropsy) that may create	Autoclave available in
		1100000	infectious aerosols	facility
			PPEs:	,
			Appropriate respiratory	
			protection	
4	Dangerous/exotic	ABSL-3 practices plus:	ABSL-3 equipment plus:	ABSL-3 facility plus:
	agents which pose	Entrance through change room	Maximum containment	Separate building or
	high risk of	1 8	equipment (i.e. Class III BSC	isolated zone
	life-threatening disease	removed and laboratory	or partial containment	Dedicated supply/exhaust,
	Aerosol	clothing is put on; shower	equipment in combination with full-body, air supplied,	vacuum, and decontamination systems
	transmission, or	upon exit • All wastes are	positive pressure personnel	Other requirements
	related agents with	decontaminated before	suit) used for all procedures	outlined in the text
	unknown risk of	removal from facility	and activities	Samuel III tile text
	transmission			

Refer to Laboratory Safety for more information regarding the use of hazardous materials with laboratory research animals. A copy of the CDC/NIH criteria for laboratory and

	Tarleton State University		
		Program:	Biological Safety
	Risk Management & Compliance	Doc. No.:	BIOS-04-L2-S0-CH0-001
		Rev No:	3
	Level 2	Date:	8-31-2018
	Level 2	Dept.:	TSU Risk Management & Compliance

animal biosafety levels is available from the Department of Risk Management and Compliance.

7. RECOMBINANT DNA RESEARCH

Tarleton State University is obligated to ensure that all recombinant DNA (rDNA) work conducted by its faculty and staff conforms to Federal rDNA guidelines. This task falls jointly to the Institutional Biosafety Committee (IBC) and the Department of Risk Management and Compliance. The IBC reviews all protocols involving rDNA, rules on the appropriateness of proposed containment procedures, and sets suitable biosafety levels. The Department of Risk Management and Compliance inspects individual laboratories and verifies that practices and facilities meet the requisite biosafety level assigned by the IBC. The Federal rDNA guidelines defines rDNA as "...(i) molecules which are constructed outside of living cells by joining natural or synthetic DNA segments to DNA molecules that can replicate in a living cell, or (ii) molecules that result from the replication of those described in (i) above." The Federal definition also includes the replicated progeny of these molecules as well as cells, plants, and animals that harbor such molecules. Transgenic plants and animals also come under the guidelines, even if the transgenic DNA was not cloned prior to introduction.

Investigators who possess rDNA in any form must file a rDNA protocol with the IBC. A copy of the TAMUS/Tarleton State University Policies and Procedures for Research Involving Recombinant DNA is available from the Department of Risk Management and Compliance.

8. DISINFECTION AND STERILIZATION

Biological safety depends on proper cleanup and removal of potentially harmful agents. Disinfection and sterilization are two ways to help ensure biological safety in the laboratory.

- a. Disinfection: Reduction of the number of pathogenic organisms by the direct application of physical or chemical agents.
- b. Sterilization: Total destruction of all living organisms.

General Guidelines for Disinfection and Sterilization Choosing the best method for disinfection and sterilization is very important. The proper method depends on the following:

i. Target organisms to be removed

	Tarleton State University		
		Program:	Biological Safety
	Risk Management & Compliance	Doc. No.:	BIOS-04-L2-S0-CH0-001
		Rev No:	3
Level 2	Lovel 2	Date:	8-31-2018
	Level 2	Dept.:	TSU Risk Management & Compliance

ii. Characteristics of the area to be cleaned

Once you have chosen the proper method for disinfection or sterilization, follow these guidelines to ensure laboratory safety:

- iii. Frequently disinfect all floors, cabinet tops, and equipment where biohazardous materials are used.
- iv. Use autoclavable or disposable materials whenever possible. Keep reusable and disposable items separate.
- v. Minimize the amount of materials and equipment used when working with infectious agents.
- vi. Sterilize or properly store all biohazardous materials at the end of each day.
- vii. Remember that some materials may interfere with chemical disinfectants-use higher concentrations or longer contact time.
- viii. Use indicators with autoclave loads to ensure sterilization.
- ix. Clearly mark all containers for biological materials (e.g., BIOHAZARDOUS TO BE AUTOCLAVED).

a. Disinfectant Factors

The effectiveness of a disinfection procedure is controlled significantly by a number of factors, each one of which may have a pronounced effect on the end result. Among these are:

- i. The nature and number of contaminating microorganisms (especially the presence of bacterial spores);
- ii. the amount of organic matter present (e.g., soil, feces, and blood);
- iii. the type and condition of instruments, devices, and materials to be disinfected:
- iv. the temperature

	Tarleton State University		
		Program:	Biological Safety
	Risk Management & Compliance	Doc. No.:	BIOS-04-L2-S0-CH0-001
		Rev No:	3
	Level 2	Date:	8-31-2018
	Level 2	Dept.:	TSU Risk Management & Compliance

Use the following table to aid in the selection of disinfectants:

Disinfectant	Uses
Alcohols	Ethyl or isopropyl alcohol at 70-80% concentration is a good general purpose surface disinfectant; not effective against bacterial spores.
Phenols	Effective against vegetative bacteria, fungi, and viruses containing lipids, unpleasant odor.
Formaldehyde	Concentration of 5-8% formalin is a good disinfectant against vegetative bacteria, spores, and viruses; known carcinogen; irritating odor.
Quaternary Ammonium Compounds	Cationic detergents are strongly surface active; extremely effective against lipoviruses; ineffective against bacterial spores; may be neutralized by anionic detergents (i.e., soaps). Works best in ph >7.
Chlorine	Low concentrations (50-500 ppm) are active against vegetative bacteria and most viruses; higher concentrations (2,500 ppm) are required for bacterial spores; corrosive to metal surfaces; must be prepared fresh; laundry bleach (5.25% chlorine) may be diluted and used as a disinfectant.
Iodine	Recommended for general use; effective against vegetative bacteria and viruses; less effective against bacterial spores; Wescodyne diluted 1 to 10 is a popular disinfectant for washing hands.

Refer to Radiation Safety for information pertaining to the use of ultraviolet lights as a method of disinfection.

b. Sterilization Methods

There are three common methods for sterilizing laboratory materials: wet heat, dry heat, and ethylene oxide gas.

i. WET HEAT - When used properly, the damp steam heat from an autoclave effectively sterilizes biohazardous waste. Sterilization occurs when contaminated materials reach 15 psi pressure at 250°F or 121°C for at least 30 minutes.

Tarleton State University		
	Program:	Biological Safety
Risk Management & Compliance	Doc. No.:	BIOS-04-L2-S0-CH0-001
	Rev No:	3
Level 2	Date:	8-31-2018
Level 2	Dept.:	TSU Risk Management & Compliance

- ii. For the autoclave process to be effective, sufficient temperature, time, and direct steam contact are essential. Every Tarleton State University department that autoclaves biohazardous waste should have written documentation to ensure the waste is sterile. Parameters for sterilization and standard operation procedures should include requirements for verifying sterilization.
- iii. Potential problems with wet heat sterilization and autoclaves include the following:
 - a) Heavy or dense loads require higher temperature for sterilization.
 - b) Poor heat conductors (e.g., plastic) take longer to sterilize.
 - c) Containers may prevent steam from reaching the materials to be sterilized.
 - d) Incomplete air removal from the chamber can prevent contact between the steam and the load:
 - Deep trays can interfere with air removal.
 - Tightly stacked loads can impede steam circulation and air removal
 - Double-bagging will impede steam penetration.
 - Carcasses do not allow steam penetration.
 - Some bags and containers rated as autoclavable have thermal stability but they do not allow steam penetration.
- iv. To ensure that all materials are sterile, always test autoclave loads. Remember, however, that some sterilization indicators are incomplete. Autoclave tape, for example, verifies sufficient external temperature exposure, but it does not indicate internal equipment temperature, exposure time, or steam penetration. Thermocouples or other instrumentation can also indicate temperature, but they do not verify sterility. A biological indicator is the most effective monitor to ensure sterility. Commercially available strips or vials of Bacillus species endospores, for example, are suitable biological indicators.

	Tarleton State University		
		Program:	Biological Safety
	Risk Management & Compliance	Doc. No.:	BIOS-04-L2-S0-CH0-001
		Rev No:	3
		Date:	8-31-2018
	Level 2	Dept.:	TSU Risk Management & Compliance

- v. DRY HEAT Less effective than wet heat for sterilizing biohazardous materials. Dry heat requires more time (two to four hours) and a higher temperature (320-338°F or 160-170°C) to achieve sterilization. A Bacillus species biological indicator can verify dry heat sterilization.
- vi. ETHYLENE OXIDE GAS lethal to all microorganisms because it is also a known carcinogen and potentially explosive (freon and carbon dioxide mixtures are stable), minimize your exposure and use extreme care when working with this gas. Ethylene oxide sterilizers and aerators must be properly vented. Ethylene oxide gas is most effective with heat-resistant organisms and heat sensitive equipment. The effectiveness of ethylene oxide gas may be affected by the following:
 - a) Temperature: The antimicrobial activity of ethylene oxide increases with increased temperature. Normal sterilization temperature is 120-140°F or 49-60°C.
 - b) Ethylene Oxide Concentration: Sterilization time decreases with increased gas concentration. Normal concentration is 500-1000 mg/L.
 - c) Humidity: Relative humidity of 30-60% is necessary.
 - d) Exposure Time: Follow the manufacturer's recommendations.

9. BIOLOGICAL SAFETY CABINETS (BSCs)

A biological safety cabinet is a primary barrier against biohazardous or infectious agents. A biological safety cabinet is often referred to by other names such as: biohood, tissue culture hood, or biological fume hood. Although biological safety cabinets surround the immediate workspace involving an agent, they do not provide complete containment (i.e., aerosols can escape). Therefore, careful work practices are essential when working with agents that require a biological safety cabinet.

All biological safety cabinets contain at least one High Efficiency Particulate Air (HEPA) filter. These cabinets operate with a laminar air flow (i.e., the air flows with uniform velocity, in one direction, along parallel flow lines.). Biological safety cabinets must be inspected and certified:

a. When newly installed

	Tarleton State University		
		Program:	Biological Safety
	Risk Management & Compliance Level 2	Doc. No.:	BIOS-04-L2-S0-CH0-001
		Rev No:	3
		Date:	8-31-2018
	Level 2	Dept.:	TSU Risk Management & Compliance

- b. After filter or motor replacement
- c. After being moved
- d. Annually

Table 3: Types of Biological Safety Cabinets (BSC)

Type of Cabinet	Operation and Use
Class I	Only exhaust air is filtered. The user and environment are protected but the experiment is not. Operator's hands and arms may be exposed to hazardous materials inside the cabinet. This cabinet may be used with low to moderate-risk biological agents.
Class II	Vertical laminar air flow with filtered supply and exhaust air. The user, product, and environment are protected.
Type A	Recirculates 70% of the air inside the cabinet. Do not use with flammable, radioactive, carcinogenic, or high-risk biological agents.
Type B1	Recirculates 30% of the air inside the cabinet and exhausts the rest to the outside. May be used with low to moderate-risk agents and small amounts of chemical carcinogens or volatiles.
Type B2	Offers total exhaust with no recirculation.
Type B3	Same as Class II Type A, but vented to the outside of the building.
Class III or	Gas-tight and maintained under negative air pressure. Used to work with highly
Glovebox	infectious, carcinogenic, or hazardous materials. All operations are conducted through rubber gloves attached to entry portals.

e. Biological Safety Cabinet Operations

Follow these guidelines for using biological safety cabinets properly:

i. Preparation

- a) Leave safety cabinets on at all times. Otherwise, turn the blower on and purge the air for at least five minutes before beginning work.
- b) Never turn off the blower of a biological safety cabinet that is vented to the outside.
- c) Turn off the UV light if it is on. Never work in a unit with the UV light illuminated. (UV light will damage your eyes.)

 Do not depend on the UV germicidal lamp to provide a sterile work surface; wipe down the surface with a disinfectant (70% alcohol is usually suitable).
- d) For more information on ultraviolet lights, refer to the Radiation Safety.
- e) Place everything needed for your procedure inside the cabinet prior to beginning work. Arrange the equipment in logical order.
- f) Provide a container for wastes inside the cabinet. (Remember, nothing should pass through the air barrier until the entire procedure is complete.)

	Tarleton State University		
		Program:	Biological Safety
	Risk Management & Compliance	Doc. No.:	BIOS-04-L2-S0-CH0-001
		Rev No:	3
		Date:	8-31-2018
	Level 2	Dept.:	TSU Risk Management & Compliance

- g) Never place any items on the air-intake grilles.
- h) Place a disinfectant-soaked towel on the work surface to contain any splatters or spills that occur.
- i) Keep the laboratory door shut and post signs stating "CABINET IN USE" on all the doors. Restrict activities that will disturb the cabinet's airflow, such as entry, egress, and walking traffic.

ii. Cabinet Use

- a) Conduct work at least four inches from the glass view panel. The middle third area is ideal.
- b) Limit arm movement and avoid motions that could disturb airflow.
- c) If a burner is necessary, use the Touch-O-Matic type with a pilot light. Since flames cause air turbulence, place burners to the rear of the workspace.
- d) Never use flammable solvents in a biological safety cabinet unless it is a total-exhaust cabinet (e.g., Class II B2).

iii. Experiment Completion

- a) Enclose or decontaminate all equipment that has been in direct contact with the infectious agent.
- b) Cover all waste containers.
- c) To purge airborne contaminants from the work area, allow the cabinet to operate for five minutes with no activity inside the cabinet.
- d) Remove all equipment from the cabinet.
- e) Decontaminate interior work surfaces.

Biological safety cabinets are not a substitute for good laboratory practices. Because aerosols can escape, take precautions to minimize aerosol production and to protect yourself from contamination.

10. CLEAN BENCHES

A clean bench has horizontal laminar air flow. The HEPA-filtered air flows across the work surface towards the operator, providing protection for the product, but no protection for the user. Because clean benches offer no protection, use a clean bench only to prepare sterile media. Do not use clean benches when working with pathogenic organisms, biological materials, chemicals, or radioactive materials.

	Tarleton State University		
		Program:	Biological Safety
4	Risk Management & Compliance	Doc. No.:	BIOS-04-L2-S0-CH0-001
		Rev No:	3
	Level 2	Date:	8-31-2018
	Level 2	Dept.:	TSU Risk Management & Compliance

11. IMPORTING AND SHIPPING BIOLOGICAL MATERIALS

The Public Health Service provides Foreign Quarantine regulations for importing etiologic agents and human disease vectors. Other regulations for packaging, labeling, and shipping, are administered jointly by the Public Health Service and the Department of Transportation. The U.S. Department of Agriculture regulates the importation and shipment of animal pathogens. It prohibits the importation, possession, and use of certain animal disease agents that pose a serious threat to domestic livestock and poultry.

12. BIOLOGICAL SPILL RESPONSE

The exact procedure for responding to a biological spill depends on the material, amount, and location of the spill. In general, follow these steps immediately after a biological spill occurs:

- a. Warn others.
- b. Leave the room; close the door.
- c. Remove contaminated garments.
- d. Wash your hands.
- e. Notify your supervisor.

Follow these steps to clean up a biological spill:

- a. Wait for any aerosols to settle.
- b. Put on protective clothing, as appropriate.
- c. Apply disinfectant to the contaminated area.
- d. Cover the area with paper towels to absorb the disinfectant.
- e. Wipe up the towels and mop the floor.
- f. Autoclave all contaminated wastes and properly dispose of wastes as required.

Spill cleanup must be appropriate for the hazards involved. Call the Department of Risk Management and Compliance for assistance.

If a spill occurs inside a biological safety cabinet, follow these steps:

- a. Decontaminate materials while the cabinet is operating to prevent contaminants from escaping.
- b. Spray or wipe all affected equipment with an appropriate disinfectant. (Wear gloves and other PPE as necessary while doing this.)

	Tarleton State University		
		Program:	Biological Safety
	Risk Management & Compliance	Doc. No.:	BIOS-04-L2-S0-CH0-001
		Rev No:	3
	Level 2	Date:	8-31-2018
	Level 2	Dept.:	TSU Risk Management & Compliance

c. If the spill is large, flood the work surface with disinfectant and allow it to stand for 10 to 15 minutes before removing it.

13. BIOLOGICAL WASTE DISPOSAL

The Texas Department of Health (TDH) and the Texas Commission on Environmental Quality (TCEQ) regulate the disposal of biohazardous waste. Waste that contains infectious materials and waste that may be harmful to humans, animals, plants, or the environment is considered biohazardous. Refer to Tarleton State University's Biohazardous Waste Program for more information.

14. BLOODBORNE PATHOGENS

Bloodborne pathogens are biological agents that cause human disease. Examples of bloodborne diseases include: Hepatitis, Syphilis, Malaria and Human Immunodeficiency Virus (HIV). Two significant and deadly bloodborne diseases are hepatitis B virus (HBV) and HIV. These pathogens may be present in the following materials:

- a. Human blood
- b. Body fluids, such as saliva, semen, vaginal secretions, phlegm, and other body fluids visibly contaminated with blood
- c. Unfixed human tissues or organs other than intact skin
- d. HIV or HBV cultures
- e. Blood, organs, or other tissues from experimental animals infected with HIV or HBV

Bloodborne pathogens may enter the body and infect you through a variety of means, including the following:

- a. Accidental injury with a sharp object contaminated with infectious material.
- b. Open cuts, nicks, and skin abrasions that come into contact with infectious materials. Other potential sites of transmission include acne sores and the mucous membranes of the mouth, nose, or eyes.
- c. Unprotected sexual activity with someone who is infected with the disease.
- d. Indirect transmission, such as touching a contaminated object and then transferring the pathogen to the mouth, eyes, nose, or open skin.

For more information, refer to the Bloodborne Pathogen Program or contact the Department of Risk Management and Compliance.

	Tarleton State University		
		Program:	Biological Safety
	Risk Management & Compliance	Doc. No.:	BIOS-04-L2-S0-CH0-001
		Rev No:	3
		Date:	8-31-2018
	Level 2	Dept.:	TSU Risk Management & Compliance

REFERENCES

Biohazardous Waste Program – Tarleton State University Bloodborne Pathogen (BBP) Program – Tarleton State University

Safe and Effective Use of the Steam Autoclave

Biosafety in Microbiological and Biomedical Laboratories – US Dept. of Health and Human Services

MSDS for Infectious Agents

Laboratory Biosafety Guidelines

CDC/NIH Primary Containment for Biohazards: Selection, Installation and Use of

Biological Safety Cabinets

Texas Health and Safety Code Regulations