

Bloodborne Pathogens and Biosafety Plan

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1. PURPOSE

The purpose of the Trinity College Biosafety Manual and Exposure Control Plan (ECP) is to explain the engineering and work practice controls, including the use of personal protective equipment, housekeeping requirements, medical surveillance, and hepatitis B vaccination programs in place to prevent and limit exposure to biohazardous materials, including human source material such as blood, serum, tissue, human cell lines and any other potentially infectious material.

This plan was developed in accordance with OSHA's Occupational Exposure to Bloodborne Pathogens; 29 CFR 1910.1030, to minimize or eliminate employee exposure to bloodborne pathogens and other biological hazards and has been formulated in accordance with the provisions of the National Institutes of Health (NIH) and the Centers for Disease Control (CDC). This manual is intended for use as a supplement to the NIH Guidelines for Research Involving rDNA Molecules and the CDC/NIH Biosafety in Microbiological and Biomedical Laboratories (BMBL).

1.1 Accessibility of the Plan

- 1.1.1 Copies of the ECP will be maintained in the following locations:
 - 1.1.1.1 With the Biological Safety Officer (BSO)
 - 1.1.1.2 Available on Trinity's EHS webpage; and
 - 1.1.1.3 Available in the Environmental Health and Safety Office (Buildings & Grounds)

1.2 Plan Review

- 1.2.1 This Laboratory Safety Plan will be reviewed annually and whenever:
 - 1.2.1.1 There are regulatory changes requiring amendments to the Plan;
 - 1.2.1.2 The list of responsible personnel changes, such as the biological safety officer;
 - 1.2.1.3 There is a change in the layout or design of the facility, new equipment, or biological materials;
 - 1.2.1.4 New types of biological materials are introduced that require special actions; and/or
 - 1.2.1.5 There are any substantial changes in operations and/or maintenance of the facility.

1.3 Definitions

- Absorption - This hazard is from direct contact, or splatters and aerosols of chemical substances. Material is absorbed through the skin or eyes and is transferred into the blood and possibly to target organs. There is also the possibility to cause occupational dermatitis or sensitization.
- Shall / Will – indicates a mandatory requirement to be followed as written.
- Should – indicates a preferred action in the execution of the policy or procedure.
- Blood – means human blood, human blood components, and products made from human blood.
- Bloodborne Pathogens - means pathogenic microorganisms that are present in human blood and can cause disease in humans. These pathogens include, but are not limited to, hepatitis B virus (HBV) and human immunodeficiency virus (HIV).

- Contaminated – means the presence or the reasonably anticipated presence of blood or other potentially infectious materials on an item or surface.
- Contaminated Laundry – means laundry which has been soiled with blood or other potentially infectious materials or may contain sharps.
- Contaminated Sharps means any contaminated object that can penetrate the skin including, but not limited to, needles, scalpels, broken glass, broken capillary tubes, and exposed ends of dental wires.
- Decontamination – means the use of physical or chemical means to remove, inactivate, or destroy bloodborne pathogens on a surface or item to the point where they are no longer capable of transmitting infectious particles and the surface or item is rendered safe for handling, use, or disposal.
- Engineering Controls – means controls (e.g., sharps disposal containers, self-sheathing needles) that isolate or remove the bloodborne pathogens hazard from the workplace.
- Exposure Incident – means a specific eye, mouth, other mucous membrane, non-intact skin, or parenteral contact with blood or other potentially infectious materials that result from the performance of an employee's duties.
- Handwashing Facilities – means a facility providing an adequate supply of running potable water, soap and single use towels or hot air-drying machines.
- Licensed Healthcare Professional – a person whose legally permitted scope of practice allows him or her to independently perform the activities required by paragraph (f) Hepatitis B Vaccination and Post-exposure Evaluation and Follow-up.
- HBV – means hepatitis B virus.
- HIV – means human immunodeficiency virus.
- Occupational Exposure – means reasonably anticipated skin, eye, mucous membrane, or parenteral contact with blood or other potentially infectious materials that may result from the performance of an employee's duties.
- Other Potentially Infectious Materials – means (1) The following human body fluids: semen, vaginal secretions, cerebrospinal fluid, synovial fluid, pleural fluid, pericardial fluid, peritoneal fluid, amniotic fluid, saliva in dental procedures, any bodily fluid that is visibly contaminated with blood, and all body fluids in situations where it is difficult or impossible to differentiate between body fluids; (2) Any unfixed tissue or organ (other than intact skin) from a human (living or dead); and (3) HIV-containing cell or tissue cultures, organ cultures, and HIV- or HBV-containing culture medium or other solutions; and blood, organs, or other tissues from experimental animals infected with HIV or HBV.
- Parenteral – means piercing mucous membranes or the skin barrier through such events as needlesticks, human bites, cuts, and abrasions.
- Personal Protective Equipment – is specialized clothing or equipment worn by an employee for protection against a hazard. General work clothes (e.g., uniforms, pants, shirts or blouses) not intended to function as protection against a hazard are not considered to be personal protective equipment.
- Recombinant DNA (rDNA) – DNA that has been formed artificially by combining constituents from different organisms.
- Regulated Waste – means liquid or semi-liquid blood or other potentially infectious materials; contaminated items that would release blood or other potentially infectious

materials in a liquid or semi-liquid state if compressed; items that are caked with dried blood or other potentially infectious materials and are capable of releasing these materials during handling; contaminated sharps; and pathological and microbiological wastes containing blood or other potentially infectious materials.

- Source Individual – means any individual, living or dead, whose blood or other potentially infectious materials may be a source of occupational exposure to the employee.
- Sterilize – means the use of a physical or chemical procedure to destroy all microbial life including highly resistant bacterial endospores.
- Universal Precautions – is an approach to infection control. According to the concept of Universal Precautions, all human blood and certain human body fluids are treated as if known to be infectious for HIV, HBV, and other bloodborne pathogens.
- Work Practice Controls – means controls that reduce the likelihood of exposure by altering the manner in which a task is performed (e.g., prohibiting recapping of needles by a two-handed technique).

2. APPLICABILITY

The provisions of the Biosafety Manual and Exposure Control Plan (ECP) apply to all personnel working in laboratories at Trinity College. This program also applies to contractors who might be exposed to biological hazards while at Trinity. Employees are encouraged to contribute to the continuous improvement of the plan with specific suggestions regarding their work or processes related to biological safety, management, and employee safety. The ECP will be reviewed annually and amended as necessary based upon the feedback provided throughout the previous year and any new recommended changes to current policies.

3. ROLES AND RESPONSIBILITIES

3.1 Biological Safety Officer/EHS Manager

- 3.1.1 Develops and implements appropriate biosafety policies and practices;
- 3.1.2 Supervises the procurement, use, and disposal of biohazardous material in the laboratory;
- 3.1.3 Conducts formal, documented, biosafety/housekeeping inspections of laboratories and safety equipment, and assures that follow-up items are addressed in a timely manner;
- 3.1.4 Assists in the design and development of safe facilities; working with Facilities and Campus Safety to assure facilities are maintained at all times;
- 3.1.5 Updates his/her knowledge relating to current legal requirements for regulated substances;
- 3.1.6 Assists Supervisors in developing policies and procedures specific to the work being conducted in their areas;
- 3.1.7 Assists Supervisors in determining which personnel require medical consultations or personal protective equipment;

- 3.1.8 Conducts accident investigations and assists in efforts to reduce the potential for recurrence of these events;
- 3.1.9 Reviews this Plan on an annual basis and making changes as needed;
- 3.1.10 Assures that adequate training programs are available and maintaining the proper documentation of that training;
- 3.1.11 Receives and reviews reports of any accident or spill that occurs in the laboratory;
- 3.1.12 Verifies that all biohazardous waste is disposed of in accordance with all municipal, state and federal regulations.

3.2 Institutional Biosafety Committee (IBC)

- 3.2.1 Approves all research involving recombinant or synthetic nucleic acid molecules
- 3.2.2 Ensures all research is conducted in accordance to NIH Guidelines
- 3.2.3 Ensures all research is being conducted with the proper controls and PPE
- 3.2.4 Creates spill-response protocol for biological materials
- 3.2.5 Meet at least annually to discuss and review current research and spill procedures
- 3.2.6 IBC members:

<u>Name</u>	<u>Committee Role</u>	<u>Date Added</u>	<u>Email address</u>
Kyle Coughlin	Biosafety Officer / EHS Representative		kjcoughlin@triumvirate.com
Robert Fleming	Chair	3/3/2014	robert.fleming@trincoll.edu
Lisa Foster	Microbiologist expert	3/3/2014	lisaanne.foster@trincoll.edu
Hebe Guardiola-Diaz	Lab representative	3/3/2014	hebe.guardioladiaz@trincoll.edu
David Ruskin	Animal expert	3/3/2014	David.ruskin@trincoll.edu
Bruce Wittchen	Local, non-affiliated	3/3/2014	bruce.wittchen@ct.gov
Mike Tortora	Local, non-affiliated	3/3/2014	mtortor@connecticutchildrens.org

3.3 Facilities Department

- 3.3.1 Ensures laboratory facility security.
- 3.3.2 Provides adequate ventilation for all laboratory needs.
- 3.3.3 Maintains the facility in compliance with the state building code.
- 3.3.4 Ensures that power, emergency power, heating, humidity, water, and lighting are maintained at adequate levels.
- 3.3.5 Ensures all air ductwork is clean and there is no breach in piping.
- 3.3.6 Ensures timely response to the laboratory facility and materials contained therein

during power outages

3.4 Lab Managers/Supervisors

- 3.4.1 Determine, with the help of the BSO/EHS manager, the appropriate levels of protective apparel and equipment for their projects.
- 3.4.2 Ensure that workers follow the ECP, including the use of the appropriate, required protective equipment and clothing.
- 3.4.3 Ensure that adequate provisions and equipment are made available for work with biohazardous materials
- 3.4.4 Ensure that this equipment is in working order, and that staff have been trained in its correct usage.
- 3.4.5 Coordinate with the Biosafety Officer to determine the level of medical surveillance appropriate for their staff and ensuring their access to it.
- 3.4.6 Assures that any/all biohazardous wastes are disposed of in accordance with all municipal, state and federal regulations.

3.5 Laboratory Employees

- 3.5.1 Participate in training programs provided by Trinity College.
- 3.5.2 Read, understand, and follow the policies and procedures outlined in the Trinity Safety Programs.
- 3.5.3 Plan and conduct each operation in accordance with the Biosafety Plan and obtain pre-approval from the BSO/EHS Manager if necessary.
- 3.5.4 Wear appropriate personal protective equipment and follow safe work practices as outlined in the Plan.
- 3.5.5 Notify Supervisors when equipment is malfunctioning, or PPE is not available.
- 3.5.6 Label and dispose of biohazardous waste in compliance with the ECP.
- 3.5.7 Notify the EHS Manager of any incident, accident or spill.

4. PRINCIPLES OF BIOSAFETY

4.1 Introduction to Biosafety

- 4.1.1 The essential elements of the four biosafety levels are designated in ascending order, by degree of protection provided to personnel, the environment, and the community.
- 4.1.2 Standard microbiological practices are common to all levels.
- 4.1.3 Special microbiological practices enhance worker safety, environmental protection, and address the risk of handling agents requiring increasing levels of containment.
- 4.1.4 **Biosafety Level 1 (BSL-1):**
 - 4.1.4.1 Biosafety Level 1 is suitable for work involving well-characterized agents not known to consistently cause disease in healthy adult humans, and present minimal potential hazard to laboratory personnel and the environment.
 - 4.1.4.2 Work is typically conducted on open bench tops using standard microbiological practices.

- 4.1.4.3 Special containment equipment or facility design is not required but may be used as determined by appropriate risk assessment.
- 4.1.4.4 Laboratory personnel must have specific training in the procedures conducted in the laboratory.

4.1.5 Biosafety Level 2 (BSL-2):

- 4.1.5.1 Biosafety Level 2 builds upon BSL-1. BSL-2 is suitable for work involving agents that pose moderate hazards to personnel and the environment.
- 4.1.5.2 It differs from BSL-1 in that:
 - 4.1.5.2.1 Laboratory personnel have specific training in handling pathogenic agents and are supervised by competent individuals in handling infectious agents and associated procedures;
 - 4.1.5.2.2 Access to the laboratory is restricted when work is being conducted.
 - 4.1.5.2.3 All procedures in which infectious aerosols or splashes may be created are conducted in BSCs or other physical containment equipment.

5. HUMAN SOURCE MATERIALS

5.1 Bloodborne Pathogens

- 5.1.1 The Occupational Safety and Health Administration (OSHA) created the Occupational Exposure to Bloodborne Pathogens Standard, 29 CFR Part 1910.1030 to minimize or eliminate exposure to infectious agents that may be present in human blood, tissues or certain body fluids.
- 5.1.2 The Bloodborne Pathogens Standard applies to all employers having employees that are “occupationally exposed” to human blood or other potentially infectious materials (OPIM).
- 5.1.3 An employee is considered occupationally exposed if there is “reasonably anticipated skin, eye, mucous membrane, or parenteral contact with human blood or other potentially infectious materials in the performance of an employee’s duties.”
- 5.1.4 Other potentially infectious materials include:
 - 5.1.4.1 Human cell or tissue cultures.
 - 5.1.4.2 Organ cultures.
 - 5.1.4.3 Any unfixed tissue or organ, other than intact skin, from a human being (living or dead);
 - 5.1.4.4 Human Immunodeficiency virus (HIV), Hepatitis B virus (HBV) or Hepatitis C virus (HCV)- containing culture media or other solutions;
 - 5.1.4.5 Human body fluids, except urine, feces, saliva or tears unless visibly contaminated with blood; and
 - 5.1.4.6 Blood, organs, or other tissues from experimental animals infected with HIV, HBV, HCV or other bloodborne pathogens.

- 5.1.5 An individual is also considered occupationally exposed if they do not have direct contact with blood or OPIM but use equipment that is used to process or store blood, OPIM, or bloodborne pathogens.
- 5.1.6 All occupationally exposed employees are required to attend a bloodborne pathogens training session prior to beginning work and annually thereafter.
- 5.1.7 OSHA has determined that occupational exposure to human blood, tissues and body fluids poses a significant health risk because they may contain bloodborne pathogens including, but not limited to:
 - 5.1.7.1 Human Immunodeficiency Virus (HIV);
 - 5.1.7.2 Hepatitis B Virus (HBV);
 - 5.1.7.3 Hepatitis C Virus (HCV);
 - 5.1.7.4 Hepatitis D Virus (HDV);
 - 5.1.7.5 Human T-lymphotropic Virus Type I;
 - 5.1.7.6 Hemorrhagic Fever Viruses; and
 - 5.1.7.7 Creutzfeldt-Jakob Virus.
- 5.1.8 The OSHA Bloodborne Pathogen Standard defines safety requirements for working with human blood and other clinical materials, HIV, and the bloodborne hepatitis viruses. Those safety requirements, also known as universal precautions, are described in this Biological Safety Manual.
- 5.1.9 Materials other than those mentioned above which should also be handled within BSL2 containment using universal precautions are:
 - 5.1.9.1 Human-derived cell lines;
 - 5.1.9.2 Human cell strains;
 - 5.1.9.3 Human serum derived reagents; and
 - 5.1.9.4 Non-human primate blood, tissues and cells.

5.2 Human Cell Lines

- 5.2.1 A Human Cell Line is defined as in vitro or animal passaged (e.g., nude mouse) cultures or human cells that fulfill traditional requirements of a cell line designation. That is, the cells are immortalized cells transformed by spontaneous mutation, natural infection, or laboratory infection with an immortalizing agent.
- 5.2.2 The following must be handled at Biosafety Level 2, within a Class II Biosafety Cabinet (BSC):
 - 5.2.2.1 All cell lines (primary and established) of human or primate origin. If cell lines are classified by other organizations as downgraded to BSL1, documentation indicating that the line is free of bloodborne pathogens is required, as described above;
 - 5.2.2.2 All cell lines from tumor tissue or transformed by any oncogenic virus or tumor suppressor;
 - 5.2.2.3 All cell lines exposed to or transformed by amphitropic packaging systems;
 - 5.2.2.4 All human clinical material (such as samples of human tissues and fluids obtained after surgical resection or autopsy);
 - 5.2.2.5 All hybridoma cell lines; and

5.2.2.6 Unknown biological systems.

5.3 Human Cell Strains

- 5.3.1 Human cell strains are defined as cells propagated in vitro from primary explants of human tissue or body fluids which have a finite lifetime (non-transformed) in tissue culture for 20-70 passages.
- 5.3.2 Human cell strains must be handled as potential biohazards unless characterized by testing to be free of bloodborne pathogens and therefore exempted from the Standard's requirements.
- 5.3.3 However, if such tissue explants or subsequent cultures are derived from human subjects known to carry bloodborne pathogens, such as hepatitis viruses or human immunodeficiency viruses, or are deliberately infected with bloodborne pathogens, they must be handled in accordance with the precautions noted in the Bloodborne Pathogens Standard.
- 5.3.4 Likewise, animal tissues, explants, or cell cultures known to be contaminated by deliberate infection with HIV or hepatitis viruses are also subject to the Standard.

5.4 Human Derived Reagents

- 5.4.1 The Centers for Disease Control cautions that all human-serum-derived reagents used in the lab, such as Human Serum Albumin (HSA), must be handled at BL2 levels with universal precautions because no test method can offer complete assurance that laboratory specimens do not contain HIV, HBV, or other infectious agents.

6. EXPOSURE DETERMINATION

6.1 Applicability

- 6.1.1 The OSHA Bloodborne Pathogens Standard requires that an exposure determination be performed in laboratories where human source materials are used.

6.2 Job Classifications

- 6.2.1 Those employees with occupational exposure to BBP or OPIM at Trinity College include any position whom has access to and, without regard to frequency, enters the laboratory space.

6.3 Tasks and Procedures

- 6.3.1 Tasks and procedures with occupational exposure at Trinity include:
 - 6.3.1.1 Work with human cell lines;
 - 6.3.1.2 Processing human tissue for assay;
 - 6.3.1.3 Generating human cell strains from explants;
 - 6.3.1.4 Shipping or receiving human source materials;
 - 6.3.1.5 Emergency First Aid response; and
 - 6.3.1.6 Closing up, boxing or moving biomedical waste containers

7. MEDICAL SURVEILLANCE, VACCINATIONS, AND POST-EXPOSURE FOLLOW-UP

7.1 Medical Surveillance

- 7.1.1 Trinity College maintains a service agreement with Hartford Medical Group in Hartford, Connecticut, which laboratory employees should use for medical surveillance and consultations in the event of occupational injuries.
- 7.1.2 Human Resources maintains medical records for each employee including:
 - 7.1.2.1 Employee's name and social security number;
 - 7.1.2.2 Employee's hepatitis B vaccination status, including vaccination dates and any medical records related to the employee's ability to receive vaccination;
 - 7.1.2.3 Results of examinations, medical testing, post-exposure evaluation and follow-up procedures;
 - 7.1.2.4 Written opinions of healthcare professionals; and
 - 7.1.2.5 Copies of information provided to healthcare professionals.
- 7.1.3 Trinity College offers medical consultation at no cost to their personnel under the following circumstances:
 - 7.1.3.1 Whenever an employee develops signs or symptoms associated with a hazardous chemical or biological agent which the employee may have been exposed to in the lab;
 - 7.1.3.2 Following a report of an exposure to human source material; and
 - 7.1.3.3 Whenever an event occurs such as a spill, leak, or explosion, which results in the likelihood of a hazardous exposure biological material.

7.2 Hepatitis B Vaccinations

- 7.2.1 The OSHA Bloodborne Pathogen Standard requires that all personnel with the potential for occupational exposure to bloodborne pathogens and other human source potentially infectious materials be offered the Hepatitis B vaccine.
- 7.2.2 Trinity College offers the Hepatitis B vaccination series to all personnel who have the potential for occupational exposure and extends post-exposure evaluation and follow-up to all personnel who have had an exposure incident.
- 7.2.3 Employees with occupational exposure at Trinity are required to submit the Hepatitis B Declination / Consent form:
 - 7.2.3.1 See Appendix B for the form
 - 7.2.3.2 If you choose to decline the vaccination:
 - 7.2.3.2.1 Check the box under "DECLINATION"
 - 7.2.3.2.2 Carefully read the statement, print, sign, and date the form and return it to your supervisor or HR
 - 7.2.3.3 If you consent to the vaccination:
 - 7.2.3.3.1 Check the box under "CONSENT FOR VACCINE AND TITER"

- 7.2.3.3.2 Carefully read the statement, print, sign, and date the form and return it to your supervisor or HR
- 7.2.3.3.3 Schedule an appointment at occupational health and bring along the form
- 7.2.3.3.4 Ensure dates for shot 1, 2, 3 and titer are filled out
- 7.2.3.4 If you have already received the vaccination:
 - 7.2.3.4.1 Check the 1st box under “STATEMENT OF PREVIOUS IMMUNIZATION”
 - 7.2.3.4.2 Include the dates you received the shots, if known
 - 7.2.3.4.3 Carefully read the statement, print, sign, and date the form and return it to your supervisor or HR
- 7.2.3.5 If you have already received the vaccination, but would like to have the titer evaluated and booster administered:
 - 7.2.3.5.1 Check the 1st and 2nd boxes under “STATEMENT OF PREVIOUS IMMUNIZATION”
 - 7.2.3.5.2 Carefully read the statement
 - 7.2.3.5.3 Print, sign, and date the form and return it to your supervisor
 - 7.2.3.5.4 Schedule an appointment at occupational health and fill out the titer and booster sections as necessary

7.3 Post-Exposure Evaluation and Follow-Up

- 7.3.1 Following a report of an exposure incident, Trinity will provide a confidential medical evaluation and follow-up, including at least the following elements:
 - 7.3.1.1 Documentation of the route(s) of exposure, and the circumstances under which the exposure incident occurred
 - 7.3.1.1.1 Appendix C is an exposure incident form (Trinity Incident/Accident Report)
 - 7.3.1.2 Identification and documentation of the source individual, unless the employer can establish that identification is infeasible or prohibited by state or local law;
 - 7.3.1.3 Collection and testing of blood for HBV and HIV serological status
 - 7.3.1.3.1 The exposed employee's blood will be collected as soon as feasible and tested after consent is obtained.
 - 7.3.1.3.2 If the employee consents to baseline blood collection but does not give consent at that time for HIV serologic testing, the sample will be preserved for at least 90 days.
 - 7.3.1.3.3 If, within 90 days of the exposure incident, the employee elects to have the baseline sample tested, such testing will be done as soon as feasible.
 - 7.3.1.4 Post-exposure preventative treatment
 - 7.3.1.4.1 Post-exposure treatment that may prevent onset of the disease will be administered.
 - 7.3.1.4.2 Pre-exposure vaccination is the most effective method for preventing HBV infection.
 - 7.3.1.4.3 Hepatitis B vaccine is recommended for any previously unvaccinated employee who has a needlestick or other

percutaneous accident with a sharp instrument or mucosal exposure to blood.

7.3.1.5 Counseling by a medical professional if necessary.

7.3.1.6 Evaluation of reported illness

7.3.2 Information Provided to the healthcare professional

7.3.2.1 Trinity will ensure that the healthcare professional evaluating an employee after an exposure incident is provided the following information:

7.3.2.1.1 A copy of this regulation;

7.3.2.1.2 A description of the exposed employee's duties as they relate to the exposure incident;

7.3.2.1.3 Documentation of the route(s) of exposure and circumstances under which exposure occurred;

7.3.2.1.4 Results of the source individual's blood testing, if available; and

7.3.2.1.5 All medical records relevant to the appropriate treatment of the employee including vaccination status which are the employer's responsibility to maintain.

7.3.3 Healthcare Professional's Written Opinion Following Exposure

7.3.3.1 Trinity will obtain and provide the employee with a copy of the evaluating healthcare professional's written opinion within 15 days of the completion of the evaluation.

7.3.3.1.1 The healthcare professional's written opinion for post-exposure evaluation and follow-up will be limited to the following information:

7.3.3.1.2 That the employee has been informed of the results of the evaluation; and

7.3.3.1.3 That the employee has been told about any medical conditions resulting from exposure to blood or other potentially infectious materials which require further evaluation or treatment.

7.3.3.1.4 All other findings or diagnoses shall remain confidential and shall not be included in the written report.

7.4 Sharps Injury Log

7.4.1 Trinity College will establish and maintain a sharps injury log for the recording of percutaneous injuries from contaminated sharps.

7.4.2 The information in the log will be recorded and maintained in a manner which protects the confidentiality of the injured employee.

7.4.3 At minimum, the sharps injury log will include the following:

7.4.3.1 the type and brand of device involved in the incident;

7.4.3.2 the department or work area where the exposure incident occurred; and

7.4.3.3 an explanation of how the incident occurred.

7.4.4 The Sharps Injury Log is contained in Appendix D

8. BIOSAFETY PROCEDURES AND PRACTICES

8.1 Introduction

- 8.1.1 Good microbiological practice is important not only for safe handling of biological material but also for ensuring good experimental results. This section outlines laboratory practices to be used for Biosafety Level One (BSL-1) and Biosafety Level Two (BSL-2) laboratories at Trinity College.
- 8.1.2 The objective of physical containment is to confine the organisms containing rDNA molecules and to reduce the potential for exposure to the laboratory worker, to persons outside the laboratory, and to the environment.
- 8.1.3 Physical containment is achieved through the use of laboratory practices, containment equipment, and special laboratory design.
 - 8.1.3.1 The primary means of physical containment is provided by laboratory practices and containment equipment.
 - 8.1.3.2 Special laboratory design provides a secondary means of protection against the accidental release of organisms outside the laboratory or to the environment.

8.2 New Proposed Research

- 8.2.1 All new research regarding recombinant or synthetic nucleic acid molecules must fill out [Trinity Infectious Registration Form](#).
- 8.2.2 Form must be submitted to IBC for their review
 - 8.2.2.1 If procedures, exposure controls, and biosafety level is adequate the IBC will approve.
- 8.2.3 If research was approved as a BSL-1 safety level and the researcher wishes to investigate new vectors that will not impact current biosafety level, no further IBC approval is required.
- 8.2.4 If new vectors are introduced that will increase BSL or other impacts, IBC must be informed to review new protocols.

8.3 Labeling and Signs

- 8.3.1 All areas and equipment that contain biohazards agents must be marked with a biohazard warning label.
- 8.3.2 It must be red or orange in color with the universal biohazard symbol and lettering in black as illustrated in Appendix E
- 8.3.3 The following are examples of biohazardous materials and are required to be labeled with a Universal Biohazard symbol:
 - 8.3.3.1 Human blood, blood products, bodily fluids, and tissues
 - 8.3.3.2 Biotechnology by-product or effluent from living organisms
 - 8.3.3.3 rDNA materials
 - 8.3.3.4 Biologic agents, some of which may harbor pathogenic agents
 - 8.3.3.5 Equipment which is used with any of the above

- 8.3.3.6 Equipment in which any of the above are stored
- 8.3.3.7 Waste receptacles including sharps containers, pails, and boxes

8.4 **Biosafety Level 1 and 2 Laboratory Requirements**

- 8.4.1 BSL-1 and 2 laboratories at Trinity College adhere to the following guidelines:
 - 8.4.1.1 All employees working with BLS-1 and BLS-2 agents must receive appropriate training prior to the start of work.
 - 8.4.1.2 Access to the laboratory is limited when work is being conducted. Doors to the lab must be kept closed and access is restricted.
 - 8.4.1.3 Extreme precautions must be taken with sharps
 - 8.4.1.4 Procedures in which infectious aerosols or splashes may be created must be conducted in biological safety cabinets or other physical containment equipment.
 - 8.4.1.5 Sign are posted on the door with the universal biohazard symbol and designating the lab as a BSL-2 lab.
 - 8.4.1.6 Methods for decontamination of infectious or regulated laboratory wastes have been established and are discussed in the following sections
 - 8.4.1.7 An eyewash facility is readily available as well as a sink for washing hands

8.5 **Standard Microbial Practices**

- 8.5.1 The following standard microbiological practices shall be practiced in both BSL-1 and BSL-2 labs at Trinity College:
 - 8.5.1.1 Employees must wash their hands after they handle viable materials, after removing gloves, and before leaving the laboratory. Sinks are available in the laboratory for this purpose.
 - 8.5.1.2 Eating, drinking, smoking, handling contact lenses, and applying cosmetics are not permitted in the labs.
 - 8.5.1.2.1 Food must be stored outside the work area in cabinets or refrigerators designated and used for this purpose only.
 - 8.5.1.2.2 Avoid contact with your mouth, eyes, nose and face as much as possible
 - 8.5.1.3 Mouth pipetting is prohibited; mechanical pipetting devices are required
 - 8.5.1.4 Minimize aerosol production during all procedures by careful lab techniques to transfer materials from one container to another.
 - 8.5.1.5 Work surfaces and equipment must be decontaminated at least once a day with an appropriate disinfectant and after any spill of viable material.
 - 8.5.1.6 Employees must utilize good housekeeping practices and keep their work areas clean and organized
 - 8.5.1.7 Substitute plastic for glass where possible.
 - 8.5.1.8 Work in a biosafety cabinet as much as possible. Minimize movements inside the biosafety cabinet.
 - 8.5.1.9 BSL-2 materials, including wastes, must be handled to prevent leakage during storage, transport, handling and processing.

- 8.5.1.10 All employees are required to wear personal protective equipment as discussed in Section 8.5

8.6 **Personal Protective Equipment (PPE)**

- 8.6.1 The following are personal protective equipment (PPE) guidelines for all Trinity laboratories:
- 8.6.1.1 Lab coats
 - 8.6.1.2 Full-length pants, or full leg coverage
 - 8.6.1.3 Safety glasses at all times
 - 8.6.1.4 Closed-toe shoes
 - 8.6.1.5 Nitrile gloves
 - 8.6.1.5.1 Gloves should be changed frequently and must not be reused once removed
 - 8.6.1.6 Lab coats must be removed, and hands washed prior to entering a non-lab area.

8.7 **Sharps**

- 8.7.1 Sharps are any of the following: needles, scalpels, razors, glass Pasteur pipettes, serological pipettes, slides, cover slips, syringes, plastic tips, or anything that can easily puncture or scrape the skin.
- 8.7.2 General Sharps Guidelines:
- 8.7.2.1 Handle sharps with extreme caution
 - 8.7.2.2 Do not overfill waste containers and do not force sharps into a full container as this action often results in puncture injuries.
 - 8.7.2.3 Needles/Syringes should be disposed of immediately after use.
 - 8.7.2.4 Do NOT remove needle from syringe and dispose of as a single unit.
 - 8.7.2.5 Do NOT recap needles
 - 8.7.2.6 Do NOT bend or break needles.

8.8 **Insect and Rodent Control**

- 8.8.1 Trinity College maintains a rodent and pest control service agreement with an outside vendor.
- 8.8.2 For further information, please email facilities@trincoll.edu

9. EQUIPMENT PROCEDURES

9.1 **Biosafety Cabinets**

- 9.1.1 Biosafety cabinets are primary containment devices that are designed to provide protection for the worker and the environment, as well as provide a work environment free of contaminants.
- 9.1.2 The effectiveness of the biosafety cabinet is directly dependent on the manner in which users perform their work.
- 9.1.3 The effectiveness of the cabinet is a function of three separate directional airflows:
- 9.1.3.1 Inward from the room through the front grille

- 9.1.3.2 Downward through a HEPA filter onto the work surface
- 9.1.3.3 Out of the cabinet through an exhaust HEPA filter.
- 9.1.4 Prior to working in the BSC employees must adhere to the following guidelines:
 - 9.1.4.1 Wipe the cabinet down with an appropriate disinfectant, such as ethanol or bleach;
 - 9.1.4.2 If the BSC was not already on, run the blower for 10 -15 min. prior to beginning work;
 - 9.1.4.3 Wear gloves;
 - 9.1.4.4 Keep the vertical sash below indicated height;
 - 9.1.4.5 Avoid disrupting the air flow of the BSC;
 - 9.1.4.6 Set-up equipment and workflow pattern; dirty to clean;
 - 9.1.4.7 Keep sides and grilles clear;
 - 9.1.4.8 When finished, wipe down the BSC surfaces with an appropriate disinfectant and leave the blower running for at least 10 to 15 minutes;
 - 9.1.4.9 UV light should not be used for primary decontamination purposes; and
 - 9.1.4.10 When the hood is not being used the sash should be lowered but not completely closed and the blower can be left on.
- 9.1.5 Turbulence in a biosafety cabinet may cause aerosols to cross-contaminate open vessels and/or escape the hood. Therefore, the following must be avoided when working in BSC:
 - 9.1.5.1 Blocking air flow grilles;
 - 9.1.5.2 Open flames;
 - 9.1.5.3 Rapid movement of arms in/out of cabinet;
 - 9.1.5.4 High traffic; and
 - 9.1.5.5 Cross drafts from doors.
- 9.1.6 The HEPA filter within the BSC only protects against particulates. The pressure gauge on the side of the cabinet indicates the performance of the filter. This gauge should be monitored daily.
 - 9.1.6.1 Contact Facilities or the Biosafety Officer if the needle deflects over to one side or the other.
- 9.1.7 Some additional requirements on use of BSCs:
 - 9.1.7.1 Do not store anything on top of the cabinet.
 - 9.1.7.2 Hazardous chemicals shall not to be used in the re-circulating air BSCs.
 - 9.1.7.3 Biohazard labels and biosafety level must be posted on the BSC.
 - 9.1.7.4 Biosafety cabinets must be tested and certified annually.
 - 9.1.7.4.1 Facilities is responsible for the testing and maintenance of biosafety cabinets
 - 9.1.7.5 If the BSC alarm goes off, cap tubes, close sash, and put a “Do Not Enter” sign on the door. Contact the Biosafety Officer immediately.

9.2 Centrifugation

- 9.2.1 Centrifugation of materials shall be done in screw cap or pressure seal tubes/bottles.
 - 9.2.1.1 These should be inverted carefully after filling to check the seal.

- 9.2.2 Aerosol-proof rotors must be used on centrifuges.
- 9.2.3 If there has been any possibility of leakage, the inner walls of the centrifuge chamber and the rotor are to be immediately decontaminated with an appropriate disinfectant.
- 9.2.4 All centrifuges must be labeled with the biohazard symbol

9.3 Growth Chambers and Shakers

- 9.3.1 All growth chambers and shakers are to be covered.
- 9.3.2 If shaking water baths are used, use copper sulfate (enough to give blue color) to prevent growth of microorganisms and mold. NEVER USE SODIUM AZIDE for this purpose.
- 9.3.3 Plastic flasks and bottles should be used whenever possible to avoid breakage.
- 9.3.4 Cotton plugged flasks are not considered open vessels as long as the plugs fit tightly, with no tendency to pop out.
- 9.3.5 All growth chambers and shakers must be labeled with the biohazard symbol.

9.4 Blenders

- 9.4.1 Safety blenders, which have been designed to contain aerosols, are available and should be used whenever possible.
- 9.4.2 Use of blenders must be completed inside a biological safety cabinet.
- 9.4.3 All blenders must be labeled with the biohazard symbol.

9.5 Aspiration

- 9.5.1 During aspiration, protect the vacuum lines by setting up a collection flask followed by an overflow flask if set-up is not at eye level.
- 9.5.2 A vacu-shield or alternate should also be placed in the line to reduce the risk of contaminating the in-house vacuum.
- 9.5.3 The aspiration set-up should have a premeasured disinfectant in the flask, which should be labeled accordingly.
- 9.5.4 If the flask is on the floor, protect against breakage with the use of secondary containment.
- 9.5.5 Work in the Biosafety Cabinet when removing the stopper from the flask.
- 9.5.6 Proper set-up for an aspiration process is illustrated below and in Appendix F.

10. DECONTAMINATION

10.1 General

- 10.1.1 Laboratories are subject to contamination by infectious and non-infectious biological material. Frequent decontamination is necessary to provide a work area that is suitable for good microbiological practices and to render contaminated material safe for handling. This section will discuss the procedures for the 3 types of decontamination.
- 10.1.2 All employees must be familiar with proper decontamination procedures at Trinity College.

10.2 Sterilization

10.2.1 Any item, device, or solution is considered to be sterile when it is completely free of all living microorganisms and viruses. Sterilization is used to decontaminate items with steam or gas.

10.2.1.1 Examples of these sterilization methods are steam sterilizers or ethylene oxide autoclaves.

10.2.2 Autoclave procedure

10.2.2.1 Before using the autoclave, check to make sure no items were left inside by the previous user that could pose a hazard.

10.2.2.2 Clean the drain strainer before loading the autoclave

10.2.2.3 Load the autoclave properly as per manufacturer's recommendations.

10.2.2.4 Before loading containers of liquids into the autoclave, the caps must be loosened to avoid having the bottles shatter during pressurization.

10.2.2.5 Individual glassware pieces should be in heat resistant plastic trays on a shelf or rack and never placed directly on the autoclave bottom or floor.

10.2.2.6 Use a tray with a solid bottom and walls to contain the contents and catch spills.

10.2.2.7 Add ¼ to ½ inch of water to the tray so the bottles will heat evenly.

10.2.2.8 Make sure plastic materials are compatible with being autoclaved.

10.2.2.9 Make sure the autoclave door is fully closed and latched and the correct cycle is selected before starting the cycle.

10.2.2.10 Wear heat resistant gloves when operating the autoclave door after a cycle.

10.2.2.11 If the door must be opened prior to the "cool down" cycle being completed, stand behind door when opening and beware rush of steam. Be sure to wear eye and face protection.

10.2.2.12 For non-liquid glassware loads allow the material to cool for 15 minutes prior to touching it with ungloved hands. If the material is waste wear at least latex or equivalent gloves to place the waste in the proper medical waste container.

10.2.2.13 For liquid loads allow the material to cool for one (1) hour before touching with ungloved hands. Inform others in the area that a heat hazard is present.

10.2.2.14 When removing items from the autoclave, wear heat resistant gloves. A rubber apron is also recommended.

10.3 Disinfection

10.3.1 Disinfection is the process of using antimicrobial agents on inanimate objects to destroy all non-spore forming organisms that could pose a hazard to humans or compromise an experiment. Usually disinfection is performed with a chemical agent, but heat can also be a type of disinfection treatment for liquid materials.

10.3.2 There are many types of chemical disinfectants used in laboratories:

10.3.2.1 **Chlorine based compounds, usually sodium hypochlorite solution (Bleach)**

- 10.3.2.1.1 This halogen is a universal decontaminant active against many microorganisms, including bacterial spores. Chlorine combines with protein and rapidly decreases in concentration in the presence of protein. Free available chlorine is the active element. It is a strong oxidizing agent and corrosive to metals. Chlorine solutions must be prepared frequently. Sodium hypochlorite is usually used as a base for chlorine decontaminants. An excellent decontaminant can be prepared from household or laundry bleach. These bleaches usually contain 5.25%, or 52,500 ppm, available chlorine. If diluted 1 to 100, the resulting solution will contain 525 ppm of available chlorine, and, if a nonionic detergent is added in a concentration of about 0.7%, a very good decontaminant is created.
- 10.3.2.2 **Alcohols, typically ethanol or isopropanol**
- 10.3.2.2.1 Ethyl or isopropyl alcohol in a concentration of 70-85% by weight is often used; however, both lose effectiveness at concentrations below 50% and above 90%. Alcohols denature proteins at somewhat slow rates in germicidal action. However, alcohols are effective decontaminants against lipid-containing viruses. A contact time of ten minutes is generally employed in efficacy tests with disinfectants. Due to the high evaporation rate of alcohols, repeated applications may be required to achieve the required ten-minute contact time for decontamination. Because of this, the OSHA Bloodborne Pathogens Standard does not recognize alcohol as an effective decontaminant for surfaces. Isopropyl alcohol is generally more effective against vegetative bacteria; ethyl alcohol is a more viricidal agent.
- 10.3.2.3 **Formaldehyde solutions**
- 10.3.2.3.1 Formaldehyde for use as a decontaminant is usually marketed as a solution of about 37% concentration referred to as formalin, or as a solid polymerized compound called paraformaldehyde. Formaldehyde in a concentration of 5% active ingredient is an effective liquid decontaminant. It loses considerable activity at refrigeration temperatures, and the pungent, irritating odors make formaldehyde solutions difficult to use in the laboratory. Formaldehyde vapor generated from solution is an effective space decontaminant for buildings or rooms, but in the vapor state in the presence of water tends to polymerize on surfaces to form paraformaldehyde, which is persistent and unpleasant. Heating paraformaldehyde to depolymerize it can liberate formaldehyde gas. In the absence of high moisture content in the air, formaldehyde released in the gaseous state forms less polymerized residues on surfaces and less time is required to

clear treated areas of fumes than is the case in the vapor state

10.3.2.4 **Iodophors, such as iodine**

10.3.2.4.1 The characteristics of chlorine and iodine are similar. One of the most popular groups of decontaminants for laboratory use are the iodophors, with Wescodyne being perhaps the most widely used. The range of dilution of Wescodyne recommended by the manufacturer is 1 oz. in 5 gal. of water (25 ppm available iodine) to 3 oz. in 5 gal. of water (75 ppm available iodine). The small amount of free iodine available in this range can rapidly be taken up by extraneous protein that may be present. Clean surfaces or clear water can be effectively treated with 75-ppm available iodine, but difficulties may be experienced if any appreciable amount of protein is present. For iodophors such as Wescodyne, it is critical that the manufacturer's written instructions are followed. Higher concentrations of iodophores are actually less effective, as the iodine is bound to itself or the carrier molecule. For washing the hands or for use as a sporicide, it is recommended that Wescodyne be diluted 1 to 10 in 50% ethyl alcohol (a reasonably good decontaminant itself.) This will give 1,600 ppm of available iodine, at which concentration relatively rapid inactivation of any and all microorganisms will occur.

10.3.2.5 **Phenol based solutions**

10.3.2.5.1 Phenol itself is not often used as a decontaminant. The odor is somewhat unpleasant and a sticky, gummy residue remains on treated surfaces. This is especially true during steam sterilization. Although phenol itself may not be in widespread use, phenol homologs and phenolic compounds are basic to a number of popular decontaminants. Phenolic compounds are effective decontaminants against some viruses, fungi, and vegetative bacteria, including rickettsiae. Phenolics are not effective in ordinary use against bacterial spores.

10.3.2.6 **Quaternary ammonium compounds**

10.3.2.6.1 After 40 years of testing and use, there is still considerable controversy about the efficacy of the Quats as decontaminants. These cationic detergents are strongly surface-active and are effective against lipid-containing viruses. The Quats will attach to proteins therefore dilute solutions will quickly lose effectiveness in the presence of proteins. Quats tend to clump microorganisms and are neutralized by anionic detergents such as soap. They have the advantages of being nontoxic, odorless, stable, non-staining, non-corrosive to metals, and inexpensive.

- 10.3.3 There is no universal disinfectant for all microbial agents. Some disinfectants are useful against many different types of microbes, others are used for very specific situations and agents. A risk assessment is performed for all agents in use to determine which disinfectant is effective against the agent in question.

10.4 Disinfectants for Work Surfaces and Reusable Items

- 10.4.1 The following disinfectants are acceptable for work surfaces and reusable items at Trinity:
- 10.4.1.1 10% solution of bleach.
 - 10.4.1.1.1 The shelf life of diluted bleach is only a few hours, so bleach should be diluted fresh immediately before use, or daily at a minimum.
 - 10.4.1.1.2 Allow a contact time of at least 10 minutes.
 - 10.4.1.2 70% solution of ethyl alcohol & isopropyl alcohol.
 - 10.4.1.2.1 The shelf life of diluted alcohol is one month.
 - 10.4.1.2.2 Allow a contact time of at least 10 minutes.
 - 10.4.1.3 Wescodyne solution made in accordance with product label.
 - 10.4.1.3.1 Allow a contact time of at least 10 minutes or more.
- 10.4.2 The following disinfectants are approved for liquid waste decontamination at Trinity:
- 10.4.2.1 Bleach to a final concentration of 10% in liquid waste.
 - 10.4.2.1.1 This disinfectant is best for human source materials.
 - 10.4.2.1.2 If the protein load is high in the liquid waste that is being disinfected, a 20% final concentration is necessary.
 - 10.4.2.1.3 Allow a contact time of at least 20 minutes.
 - 10.4.2.2 Wescodyne to a 1.0% final concentration in liquid waste.
 - 10.4.2.2.1 Wescodyne is an iodoform disinfectant/detergent.
 - 10.4.2.2.2 This is acceptable for tissue culture media and other such solutions but not for any human source materials.
 - 10.4.2.2.3 Allow a contact time of at least 20 minutes.
- 10.4.3 The following Disinfectants are approved for spill clean-up:
- 10.4.3.1 Bleach concentration of 10 to 20%
 - 10.4.3.2 A 70% ethanol rinse solution can follow bleach when cleaning up a spill.
 - 10.4.3.3 Approximately twice the volume of disinfectant to the volume of the spill should be used.
 - 10.4.3.4 Allow a contact time of at least 30 minutes.

11. WASTE MANAGEMENT

11.1 Liquid Biological Waste

- 11.1.1 Decontamination of liquid biological waste takes place in the biological safety

cabinet.

- 11.1.2 Cell culture waste and other liquid waste are emptied into a container which contains disinfectant.
- 11.1.3 If using bleach as the disinfectant, use 10% final concentration in the liquid volume.
- 11.1.4 The waste must remain in contact with the disinfectant for at least 30 minutes.
- 11.1.5 The disinfected liquid may then be carefully transported to the appropriate sink for disposal into the sink drain.
- 11.1.6 See Appendix G for specific steps for the bleach inactivation of liquid biological waste.

11.2 **Solid Biological Waste**

- 11.2.1 All non-sharps equipment and supplies, which have been in direct contact with biological materials, such as gloves, bench paper, plastic ware, and culture plates should be placed in a leak proof, labeled, covered container that is lined with a biohazard bag.
- 11.2.2 Each lab will have a red biohazardous step-can as well as a smaller container on the countertops for pipette tips in which to collect solid biological waste
- 11.2.3 All biological material is to be autoclaved. Once the bags have been autoclaved, they can be placed in the trash.
- 11.2.4 Sharps must be segregated from other biological or biohazardous waste
- 11.2.5 When finished using a sharp, it must be discarded immediately into a container that is:
 - 11.2.5.1 Closeable ;
 - 11.2.5.2 Leakproof (in good condition), i.e. not broken;
 - 11.2.5.3 Rigid, puncture-resistant, shatterproof; and
 - 11.2.5.4 Appropriately labeled with the biohazard symbol.
 - 11.2.5.5 Reference Appendix J for a sharp's container guide.
- 11.2.6 Full sharps containers will be removed by either the Lab Manager or EHS Manager. Full containers will be brought to the Health Center for proper disposal through a licensed contractor.
- 11.2.7 Never place your hands inside of a sharp's container.
- 11.2.8 Non-hazardous waste and containers thereof are discouraged from being discarded in sharps containers unless they meet the definition of a sharp (e.g. broken glass bottle with liquid remaining inside)

12. EMERGENCY PROCEDURES

12.1 **Emergency Spill Procedure**

- 12.1.1 In the event of an emergency spill:
 - 12.1.1.1 Inform coworkers of the situation;
 - 12.1.1.2 Contain the spill if it is safe to do so;
 - 12.1.1.3 Evacuate yourself and others from the area;
 - 12.1.1.4 Restrict access to the spill. Immediately contact the EHS Manager and the BSO; and

- 12.1.1.5 Follow the emergency evacuation procedures as outlined in the Emergency Action Plan if necessary

12.2 **Spill Supplies**

- 12.2.1 Spill kits are located throughout the labs at Trinity and contain the supplies required for the containment and clean-up of small, incidental spills in the labs.
- 12.2.2 Employees should be thoroughly familiar with the materials contained in the spill kits.
- 12.2.3 If you use a spill kit, an incident report must be completed, and the EHS manager and chemical hygiene officer must be notified.
- 12.2.4 Contents include:
 - 12.2.4.1 Caution door sign or caution tape
 - 12.2.4.2 Universal absorbent pads
 - 12.2.4.3 Booms, pillows
 - 12.2.4.4 Scoop and scraper
 - 12.2.4.5 Bags for disposal
 - 12.2.4.6 Goggles
 - 12.2.4.7 Nitrile/rubber gloves
 - 12.2.4.8 Face masks/booties
 - 12.2.4.9 Additionally, disinfectant and paper towels are located throughout the labs for use in case of a biological spill incident.

12.3 **Biohazardous Spill in a Biosafety Cabinet**

- 12.3.1 Chemical decontamination procedures should be initiated at once while the cabinet continues to operate to prevent escape of contaminants from the cabinet.
 - 12.3.1.1 Spray or wipe walls, work surfaces and equipment with an approved disinfectant
 - 12.3.1.2 Flood the top work surface tray, and the drain pans and catch basins below the work surface, with disinfectant and allow to stand for 30 minutes.
 - 12.3.1.3 Remove excess disinfectant from the tray by wiping with a sponge or cloth soaked in disinfectant.

12.4 **Biohazardous Spill Outside of a Biosafety Cabinet**

- 12.4.1 The BSO must be notified immediately when there is a spill of more than 250 ml. of viable BSL-1 organisms or more than 100 ml. of viable BSL-2 organisms outside of a biosafety cabinet
- 12.4.2 Employees must adhere to the following procedure in the event of a spill:
 - 12.4.2.1 Leave the room immediately, making sure to evacuate all others from the room as well, and close the door.
 - 12.4.2.2 Warn others not to enter the contaminated area.
 - 12.4.2.3 Remove and put any contaminated garments into a container for biohazardous waste disposal.
 - 12.4.2.4 Thoroughly wash hands and face.

- 12.4.2.5 Wait 30 minutes to allow dissipation of aerosols created by the spill.
- 12.4.2.6 Put on a long-sleeved gown, surgical mask and rubber gloves before reentering the room.
- 12.4.2.7 Pour a decontaminant solution around the spill and allow to flow into the spill.
- 12.4.2.8 Paper towels soaked in the decontaminant may be used to cover the area.
- 12.4.2.9 To minimize the generation of aerosols, avoid pouring the decontaminant solution directly onto the spill.
- 12.4.2.10 Let stand for 30 minutes to allow for an adequate contact time.
- 12.4.2.11 Using a dustpan and squeegee, transfer all contaminated materials into a biohazard bag.
- 12.4.2.12 Handle per biohazard waste procedures.

12.5 **Exposure Response**

- 12.5.1 Immediate response to a biological exposure is necessary to prevent possible infection.
- 12.5.2 If an exposure to a biological material occurs, it is important to identify the material immediately and obtain a sample for evaluation if it has not been previously tested.
- 12.5.3 If testing data is available in the safety records or from the supplier, obtain the results immediately.
- 12.5.4 **General**
 - 12.5.4.1 All overt exposures from parenteral inoculation and/or exposure to mucous membranes **MUST** be reported immediately to the supervisor and biological safety officer.
 - 12.5.4.2 Medical attention must be promptly sought for any exposures of this type, but especially for exposures to human source materials.
 - 12.5.4.3 Counseling regarding the risk of infection may be given by a medical professional, either at the emergency room, or Occupational Health Center.
 - 12.5.4.4 All exposures must be reported through an incident report and sent to the EHS office within 24 hours.
- 12.5.5 **Exposure to the Eyes/Face**
 - 12.5.5.1 Proceed to the closest eyewash or have someone guide you there
 - 12.5.5.2 Turn the flow of water on by pushing the lever or have someone turn it on for you
 - 12.5.5.3 Employees should use the saline wash bottles prior to the eyewash to ensure adequate flushing of the system
 - 12.5.5.4 Open your eyelids with your thumb and index finger
 - 12.5.5.5 Begin flushing your eyes and continue to do so for at least 15 minutes
- 12.5.6 **Needlestick/Skin Exposure**

- 12.5.6.1 Proceed to the closest sink or have someone guide you there
 - 12.5.6.2 Thoroughly wash hands for a minimum of 15 minutes with an antibacterial soap
 - 12.5.6.3 Immediately report the incident and proceed to Occupational Health or the Emergency Room, if necessary
- 12.5.7 Open Wound Exposure
- 12.5.7.1 Proceed to the closest sink or have someone guide you there
 - 12.5.7.2 Thoroughly wash exposed area for a minimum of 15 minutes with an antibacterial soap
 - 12.5.7.3 Immediately report the incident and proceed to Occupational Health or the Emergency Room, if necessary
- 12.5.8 Mucous Membrane Exposure
- 12.5.8.1 Proceed to the closest sink or have someone guide you there
 - 12.5.8.2 Thoroughly wash exposed area for a minimum of 15 minutes with an antibacterial soap
 - 12.5.8.3 Immediately report the incident and proceed to Occupational Health or the Emergency Room, if necessary
- 12.5.9 Exposure to Clothing
- 12.5.9.1 Proceed to the closest sink or have someone guide you there
 - 12.5.9.2 Thoroughly wash exposed area for a minimum of 15 minutes with an antibacterial soap
 - 12.5.9.3 Immediately report the incident and proceed to Occupational Health or the Emergency Room, if necessary

13. TRAINING

13.1 Applicability

- 13.1.1 Trinity College will ensure that all employees with occupational exposure participate in a training program which must be provided at no cost to the employee and during working hours.
- 13.1.2 Training will be provided as follows:
 - 13.1.2.1 At the time of initial assignment to tasks where occupational exposure may take place; and at least annually thereafter; and
 - 13.1.2.2 Annual training for all employees will be provided within one year of their previous training.
 - 13.1.2.3 The college will provide additional training when changes such as modification of tasks and procedures or the institution of new tasks and procedures affect the employee's occupational exposure. The additional training may be limited to addressing the new exposures created.

- 13.1.2.4 The person conducting the training shall be knowledgeable in the subject matter covered by the elements contained in the training program as it relates to the workplace that the training will address.

13.2 Training Content

- 13.2.1 An accessible copy of the regulatory text of this standard and an explanation of its contents;
- 13.2.2 A general explanation of the epidemiology and symptoms of bloodborne diseases;
- 13.2.3 An explanation of the modes of transmission of bloodborne pathogens;
- 13.2.4 An explanation of the employer's exposure control plan and the means by which the employee can obtain a copy of the written plan;
- 13.2.5 An explanation of the appropriate methods for recognizing tasks and other activities that may involve exposure to blood and other potentially infectious materials;
- 13.2.6 An explanation of the use and limitations of methods that will prevent or reduce exposure including appropriate engineering controls, work practices, and personal protective equipment;
- 13.2.7 Information on the types, proper use, location, removal, handling, decontamination and disposal of personal protective equipment;
- 13.2.8 An explanation of the basis for selection of personal protective equipment;
- 13.2.9 Information on the hepatitis B vaccine, including information on its efficacy, safety, method of administration, the benefits of being vaccinated, and that the vaccine and vaccination will be offered free of charge;
- 13.2.10 Information on the appropriate actions to take and persons to contact in an emergency involving blood or other potentially infectious materials;
- 13.2.11 An explanation of the procedure to follow if an exposure incident occurs, including the method of reporting the incident and the medical follow-up that will be made available;
- 13.2.12 Information on the post-exposure evaluation and follow-up that the employer is required to provide for the employee following an exposure incident;
- 13.2.13 An explanation of the signs and labels and/or color coding required by paragraph
- 13.2.14 An opportunity for interactive questions and answers with the person conducting the training session.

13.3 Training Records

- 13.3.1 Training records will include the following information:
 - 13.3.1.1 The dates of the training sessions;
 - 13.3.1.2 The contents or a summary of the training sessions;
 - 13.3.1.3 The names and qualifications of persons conducting the training; and
 - 13.3.1.4 The names and job titles of all persons attending the training sessions.
- 13.3.2 Training records will be maintained for 3 years from the date on which the training occurred and will be provided upon request.

14. DOCUMENT HISTORY

Document Name	Document Number	Revision Number	Description	Effective Date
Bloodborne Pathogens and Biosafety Plan		0.0	New Document	

15. APPENDICES

DOCUMENT NAME	DOCUMENT NUMBER	REVISION NUMBER
BBP and Biosafety		0.0

Appendix A – Exposure Determination Matrix

Job	Laboratory Employees	Facilities/ Shipping + Receiving	Emergency 1 st Aid Responders
Task			
Work with Human Cell Lines	X		
Processing Human Tissue for Assay	X		
Generating Human Cell Strains from Explants	X		
Shipping or Receiving Human Source Material	X	X	
Providing 1st Aid			X
Handling Biological Waste	X	X	X

Appendix B – Hepatitis B Vaccination Declination / Consent Form

Check and sign **one** of the following boxes:

Declination	
<input type="checkbox"/>	I understand that due to my occupational exposure to blood or other potentially infectious materials I may be at risk of acquiring a hepatitis B virus (HBV) infection. I have been given the opportunity to be vaccinated with the hepatitis B vaccine, at no charge to myself.

However, I decline hepatitis B vaccination at this time. I understand that by declining this vaccine, I continue to be at risk of acquiring hepatitis B, a serious disease. If in the future I continue to have occupational exposure to blood or other potentially infectious materials and I want to be vaccinated with the hepatitis B vaccine, I can receive the vaccination series at no charge to me.

Name: _____ Signature: _____

Date: _____

Consent for Vaccine & Titer

- I understand that due to my occupational exposure to blood or other potentially infectious materials I may be at risk of acquiring a hepatitis B virus (HBV) infection. I understand the risks and benefits of the hepatitis B vaccine and that I will need to receive a series of three shots followed by a scheduled titer to complete the vaccine. I would like to participate in the hepatitis B vaccination program as offered. The vaccination series and titer are offered at no cost to me. I agree to go to **[INSERT PROVIDER NAME AND ADDRESS]** to participate in this program.

Name: _____ Signature: _____

Date: _____

Shot 1 Date: _____ Shot 2 Date: _____ Shot 3 Date: _____

Consent for Titer Only

- I have previously received the hepatitis B vaccination or have acquired immunity and would like to have my titer evaluated and a booster administered if necessary.

Name: _____ Signature: _____

Date: _____

Titer Date: _____ Booster Date (if necessary): _____

DOCUMENT NAME	DOCUMENT NUMBER	REVISION NUMBER
BBP and Biosafety		0.0

Appendix C – Exposure Report Form

Refer to Trinity College Incident and Accident Reporting Form

Laboratory Incident Report: Trinity College

Please report all injuries, spills, or other chemical or equipment related incidents within 3 days to a Chemical Hygiene Officer.

Chemical Hygiene Staff:

CHO	Department	Extension
Erin Moesteller	Biology Department	2227
Jim McClean	Chemistry Department	2512
Kyle Coughlin	B&G – EH&S Manager	4250

Person reporting incident: _____

Office: _____ Phone: _____

Date of Incident: _____

Location of Incident: _____

Type of Incident: _____

Description of Events: _____

Actions Taken: _____

Notifications Made:

Campus Safety _____

CHO _____

Medical Office _____

Follow up: _____

Date report received by CHO: _____

DOCUMENT NAME	DOCUMENT NUMBER	REVISION NUMBER
BBP and Biosafety		0.0

Appendix D – Sharps Injury Log

Date	Case/Report Number	Type of Device (syringe, box cutter)	Brand Name of Device	Work Area Where Injury Occurred	Incident Description

29 CFR 1910.1030, OSHA’s Bloodborne Pathogens Standard, in paragraph (h)(5), requires an employer to establish and maintain a Sharps Injury Log for recording all percutaneous injuries in a facility occurring from contaminated sharps. The purpose of the Log is to aid in the evaluation of devices being used in healthcare and other facilities and to identify problem devices or procedures requiring additional attention or review. This log must be kept in addition to the injury and illness log required by 29 CFR 1904. The Sharps Injury Log should include all sharps injuries occurring in a calendar year. The log must be retained for five years following the end of the year to which it relates. The Log must be kept in a manner that preserves the confidentiality of the affected employee.

DOCUMENT NAME	DOCUMENT NUMBER	REVISION NUMBER
BBP and Biosafety		0.0

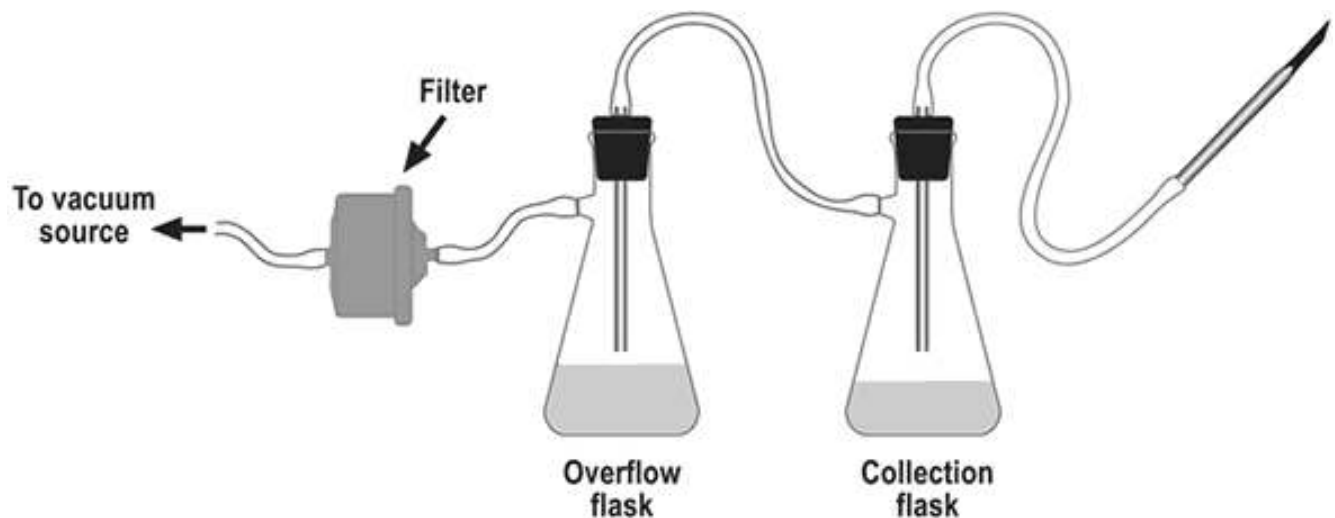
Appendix E – Biohazard Symbol



DOCUMENT NAME	DOCUMENT NUMBER	REVISION NUMBER
BBP and Biosafety		0.0

Appendix F – Aspiration Filter Setup

1. Protect the vacuum lines by setting up a collection flask followed by an overflow flask. If the flasks are on the floor, protect against breakage with the use of secondary containment.
2. A vacu-shield or alternate should also be placed in the line to reduce the risk of contaminating the in-house vacuum or vacuum pump.
3. Fill the first flask with household bleach to ~10% of the flask's volume. If a different EPA-approved disinfectant is utilized, add the volume of disinfectant required to achieve the manufacturer's recommended final concentration.
4. Aspirate the tissue culture waste into the first flask containing disinfectant. Discontinue use when the vacuum flask is 75% full.
5. Leave at room temperature for minimum of 20 minutes or let sit overnight to ensure sufficient contact time with disinfectant .disinfectant.
6. Following minimum contact time remove stopper from flask(s) inside a biosafety cabinet and dispose of contents into a laboratory sink.



Appendix G – Bleach Inactivation of Liquid Biowaste

1. Effectiveness

Bleach, a sodium hypochlorite solution, is a broad-spectrum disinfectant that is effective for enveloped viruses (e.g. HIV, HBV, HSV), vegetative bacteria (e.g. *Pseudomonas*, *Staphylococcus*, and *Salmonella*), fungi (e.g. *Candida*), mycobacterium (e.g. *M. tuberculosis*, *M. bovis*), and non-enveloped viruses (e.g. Adenovirus, Parvovirus).

Clorox bleach EPA registration number is 5813-50

2. Recommended Personal Protective Equipment for Handling Bleach and Solutions

- a. Lab coat
- b. Nitrile gloves (latex is effective as well)
- c. Safety Glasses

3. Disinfectant Concentration

The appropriate concentration of sodium hypochlorite for disinfecting liquid biological waste is 5,000 parts per million or approximately 0.5%. Household bleach is 5.2 – 6.1% sodium hypochlorite; therefore a 1:10 (V/V) dilution of bleach to liquid biological waste is appropriate.

Using Clorox (5.25% hypochlorite) in a 1:10 dilution (1-part Clorox and 9 parts liquid) yields 5,250 ppm or a 0.53% hypochlorite solution for use within 24 hours,

4. Contact Time

An appropriate contact time of sodium hypochlorite with liquid waste is 20 minutes. After a minimum of 20 minutes of contact time, disinfected liquid biological waste can be poured down the drain and flushed with water.

5. Stability and Storage

Bleach should be stored between 50 and 70° F. According to Clorox, in-diluted household bleach has a shelf life of six months to one year from date of manufacture after which bleach degrades at a rate of 20% per year.

A 1:10 bleach solution has a shelf life of 24 hours.