Bowdoin College Institutional Biosafety Committee

Recombinant DNA Research Protocol Application Form

Because NIH funds some research at Bowdoin, all research utilizing recombinant DNA conducted at or sponsored by Bowdoin must comply with the *NIH Guidelines*.
http://oba.od.nih.gov/rdna/nih_guidelines_oba.html
The Institutional Biosafety Committee (IBC) at Bowdoin is responsible for ensuring that all recombinant DNA research at Bowdoin is conducted in compliance with the *NIH Guidelines*.

Recombinant DNA as defined by NIH is either: (1) molecules that are constructed outside living cells by joining **natural or synthetic** DNA segments to DNA molecules that can replicate in a living cell, or (2) molecules that result from the replication of those described in (1) above.

IBC approval is only necessary for the use of potentially biohazardous or transgenic organisms. Recombinant DNA work done in vitro, in common laboratory E. coli strains, or transiently in organisms without stable germline transmission may be exempt under the NIH Guidelines. The first part of this application is meant to determine whether your research is exempt from the IBC process or not.

1. **Do you work with transgenic organisms or modified viruses?**

   ☐ **Yes** ☐ **No**

2. **List the host-vector systems used in your lab:**

3. **A number of systems are exempt from IBC oversight. Common examples include:**

   i) ***E. coli*** **K-12 host-vector systems (Appendix C-II)**
   ii) Saccharomyces host-vector systems (Appendix C-III)
   iii) Some select Bacillus host-vector systems (Appendix C-IV)
   If you work with additional host-vector systems you think might be exempt, please check section III-F, Appendices A, or C of *The Guidelines* to confirm the exemption. Based on the information above, do you believe your research is exempt from IBC review based on the "NIH Guidelines for Research Involving Recombinant DNA Molecules?

   ☐ **Yes**

   i. **Section:**

   ii. **Appendix:**

   ☐ **No**

4. **Do you anticipate needing containment level above BSL- 1?** (BSL-1 = Suitable for work involving well characterized agents not known to consistently cause disease in immunocompetent adult humans, and presents minimal potential hazard to laboratory personnel and the environment. Please see Appendix G for more information).

   ☐ **Yes** ☐ **No**

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5. **Do you intend to express a gene that encodes a protein that is foreign to the host (not including in *E. coli* K-12)?**

   ☐ **Yes** ☐ **No**

6. **Will this work create resistance to therapeutically useful antimicrobials or antivirals?**

   ☐ **Yes** ☐ **No**

7. **Will this work enhance the virulence of a pathogen or render a non-pathogen virulent?**

   ☐ **Yes** ☐ **No**

8. **Will this work increase the transmissibility of a pathogen?**

   ☐ **Yes** ☐ **No**

9. **Will this work alter the host range of a pathogen?**

   ☐ **Yes** ☐ **No**

10. **Will this work enhance the risk an organism may pose to the environment?** (e.g. providing herbicide resistance to a plant species.)

    ☐ **Yes** ☐ **No**

11. **Could this work enable the evasion of diagnostic/detection modalities?** (e.g., microencapsulation to avoid antibody-based detection and/or the alteration of gene sequences to avoid detection by established molecular methods)

    ☐ **Yes** ☐ **No**

12. **Will you conduct large-scale growth experiments involving an excess of 10 liters of culture?**

    ☐ **Yes** ☐ **No**

13. **Do you intend to clone any genes that encode products that are toxic for vertebrates?**

    ☐ **Yes** ☐ **No**

If you work in a non-exempt host-vector system, a transgenic organism, or if you answered "yes" to any of the questions 4-13, please complete part II of this application.

A signature below indicates that the principal investigator (PI) assumes responsibility for this project. In addition, the PI assumes responsibility for the adequate training of all personnel associated with this project.

I agree to comply with all applicable requirements pertaining to:

- Reporting of all personnel exposures and any release of rDNA material
- Reporting any organism escape, and
- Shipment/transfer of Biohazardous/ Recombinant DNA materials.

I am familiar with and agree to abide by the provisions of all IBC, CDC, OSHA, NIH or other applicable guidelines/ regulations pertaining to the proposed project. The information above is accurate and complete.

_____

Principal Investigator Date

Bowdoin College Institutional Biosafety Committee

Recombinant DNA Research Protocol Application Form
Part II

Principal Investigator Information

Name:

Department/Program:

Campus Address/ Box #:

Email:

Telephone:

14. **Description of the Project**: Please attach a brief narrative description of your proposed research with emphasis on any matters relevant to recombinant DNA and transgenic organisms. Please include enough information about your research so committee members not familiar with your work will be able to adequately evaluate the project.

15. **Transgenic Organisms:** Please describe the 1) Source species and strain; 2) the host species and strain; 3) a description of the DNA to be recombined, including the approximate size and % of genomes

16. **Biosafety Level:** Which biosafety level is adequate for your work (see descriptions below) and why? Please describe what actions you will take to maintain this level within your lab.

17. **Containment Plan:** Please provide a description of the biological and physical methods used to assure containment of recombinant materials and transgenic organisms. Will minor events such as power outages cause an accidental release? What are the potential risks of accidentally introducing this organism into the environment?

18. **Emergency Plan:** Please describe how your transgenic organisms should be handled in an emergency where your routine containment plan becomes impossible (fire, spill, accidental release etc.). Are animals to be destroyed or moved to a second location? Describe the second location and the personnel who will help and how they will be recruited.

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Biosafety level 1

This level is suitable for work involving well-characterized agents not known to consistently cause disease in healthy adult humans, and of minimal potential hazard to laboratory personnel and the environment (CDC, 1997).

It includes several kinds of bacteria and viruses including canine hepatitis, non-pathogenic *Escherichia coli*, as well as some cell cultures and non-infectious bacteria. At this level precautions against the biohazardous materials in question are minimal, most likely involving gloves and some sort of facial protection. The laboratory is not necessarily separated from the general traffic patterns in the building. Work is generally conducted on open bench tops using standard microbiological practices. Usually, contaminated materials are left in open (but separately indicated) rubbish receptacles. Decontamination procedures for this level are similar in most respects to modern precautions against everyday microorganisms (i.e., washing one's hands with anti-bacterial soap, washing all exposed surfaces of the lab with disinfectants, etc.). In a lab environment all materials used for cell and/or bacteria cultures are decontaminated via autoclave. Laboratory personnel have specific training in the procedures conducted in the laboratory and are supervised by a scientist with general training in microbiology or a related science.

Biosafety level 2

This level is similar to Biosafety Level 1 and is suitable for work involving agents of moderate potential hazard to personnel and the environment. It includes various bacteria and viruses that cause only mild disease to humans, or are difficult to contract via aerosol in a lab setting, such as *C. difficile*, hepatitis A, B, and C, influenza A, Lyme disease, dengue fever, *Salmonella*, mumps, measles, HIV,[8] scrapie, MRSA, and VRSA. Genetically modified organisms have also been classified as level 2 organisms, even if they pose no direct threat to humans. This designation is used to limit the release of modified organisms into the environment. Approval by the FDA is required to release these organisms. An example is genetically modified food crops. BSL-2 differs from BSL-1 in that:

- Laboratory personnel have specific training in handling pathogenic agents and are directed by scientists with advanced training
- Access to the laboratory is limited when work is being conducted
- Extreme precautions are taken with contaminated sharp items
- Certain procedures in which infectious aerosols or splashes may be created are conducted in biological safety cabinets or other physical containment equipment

Biosafety level 3

This level is applicable to clinical, diagnostic, teaching, research, or production facilities in which work is done with indigenous or exotic agents that may cause serious or potentially lethal disease after inhalation. It includes various bacteria,

parasites and viruses that can cause severe to fatal disease in humans, but for which vaccines or other treatment exist, such as *Leishmania donovani*, *Mycobacterium tuberculosis*, *Bacillus anthracis*, West Nile virus, Venezuelan equine encephalitis virus, Eastern equine encephalitis virus, Hendra virus, SARS coronavirus, *Salmonella typhi*, *Coxiella burnetii*, Rift Valley fever virus,

Rickettsia rickettsii, and yellow fever virus.

Laboratory personnel have specific training in handling pathogenic and potentially lethal agents, and are supervised by competent scientists who are experienced in working with these agents. This is considered a neutral or warm zone.

All procedures involving the manipulation of infectious materials are conducted within biological safety cabinets, specially designed hoods, or other physical containment devices, or by personnel wearing appropriate personal protective clothing and equipment. The laboratory has special engineering and design features.

It is recognized, however, that some existing facilities may not have all the facility features recommended for Biosafety Level 3 (i.e., double-door access zone and sealed penetrations). In this circumstance, an acceptable level of safety for the conduct of routine procedures, (e.g., diagnostic procedures involving the propagation of an agent for identification, typing, susceptibility testing, etc.), may be achieved in a biosafety level 2 (P2) facility, providing

- The filtered exhaust air from the laboratory room is discharged to the outdoors
- The ventilation to the laboratory is balanced to provide directional airflow into the room
- Access to the laboratory is restricted when work is in progress
- The recommended Standard Microbiological Practices, Special Practices, and Safety Equipment for Biosafety Level 3 are rigorously followed

The decision to implement this modification of biosafety level 3 recommendations is made only by the laboratory director.

Biosafety level 4

This level is required for work with dangerous and exotic agents that pose a high individual risk of aerosol-transmitted laboratory infections, agents which cause severe to fatal disease in humans for which vaccines or other treatments are *not* available, such as Bolivian and Argentine hemorrhagic fevers, Marburg virus, Ebola virus, Lassa fever, Crimean-Congo hemorrhagic fever, Smallpox, and other various hemorrhagic diseases. When dealing with biological hazards at this level the use of a Hazmat suit and a self-contained oxygen supply is mandatory. The entrance and exit of a Level Four biolab will contain multiple showers, a vacuum room, an ultraviolet light room, and other safety precautions designed to destroy all traces of the biohazard. Multiple airlocks are employed and are electronically secured to prevent both doors opening at the same time. All air and water service going to and coming from a biosafety level 4 (or P4) lab will undergo similar decontamination procedures to eliminate the possibility of an accidental release.

Agents with a close or identical antigenic relationship to Biosafety Level 4 agents are handled at this level until sufficient data is obtained either to confirm continued work at this level, or to work with them at a lower level.

Members of the laboratory staff have specific and thorough training in handling extremely hazardous infectious agents and they understand the primary and secondary containment functions of the standard and special practices, the containment equipment, and the laboratory design characteristics. Qualified scientists who are trained and experienced in working with these agents supervise them. The laboratory director strictly controls access to the laboratory.

The facility is either in a separate building or in a controlled area within a building, which is completely isolated from all other areas of the building. A specific facility operations manual is prepared or adopted. Building protocols for preventing contamination often use negatively pressurized facilities, which, if compromised, would severely inhibit the containment of an outbreak of aerosol pathogens.

Within work areas of the facility, all activities are confined to Class III biological safety cabinets, or Class II biological safety cabinets used with one-piece positive pressure personnel suits ventilated by a life support system. The Biosafety Level 4 laboratory has special engineering and design features to prevent microorganisms from being disseminated into the environment. The laboratory is kept at negative air pressure, so that air flows into the room if the barrier is penetrated or breached. Furthermore, an airlock is used during personnel entry and exit.