| Protocol #: |
|---|

Stevens Institute of Technology

Registration Document for Biohazards

| P.I Information | Location of Study |
|---|---|
| Name: | Building: |
| Title: | Room #s: |
| Department: | o Shared Facilities: |
| E-Mail: | o Available Containment Equip: |
| Phone: | Date: |

**Section A: General/Administrative Information**

**Introduction**

Protocol Title:

PI's Anticipated Biosafety Level:

Brief Description of Protocol (please describe experimental protocol including how the biological material will be utilized in the laboratory):

**Material Use Checklist** (Please check the materials that are used in your lab then complete the specified section for each material)

Recombinant DNA:

Genetic manipulation of microorganisms including inserting or deleting genes, use of viral vectors, development of human gene therapy, experiments involving siRNA, development of synthetic DNA constructs, etc.

| | |
|---|---|
| | Recombinant DNA, gene transfer and/or host vector systems |
| | Use of transgenic animals (including knockouts, knock ins, crossbreeding of two different transgenic strains) |
| | Use of transgenic plants |
| | Complete Section B for Exempt rDNA Experiments |
| | Complete Section C for Non-Exempt rDNA Experiments |

Potentially Infectious Microorganisms/Infectious agents:

| | |
|---|---|
| | Bacteria |
| | Virus |
| | Yeast and other Fungi |
| | Prions and/or Parasitic Agents |
| | Complete Section D for Potentially Infectious Biological Agents |
| | Complete Section D for Host Organisms Listed in Section B and C (Above) |

Human/Non-Human Primate Derived Materials, Blood, Body Fluids, Cell Lines:

| | |
|---|---|
| | Human cell lines including established human cell lines |
| | Primary human tissue explants |
| | Non-Human primate cell lines |
| | Primary non-human primate tissue explants |
| | Human non-human primate blood, body fluids |
| | Complete Section E for human and non-human primate cell lines and tissue explants |

Other:

| | |
|---|---|
| | Biological Toxins - NOT Select Agents (please complete section F) |
| | CDC/APHIS Select Agents |
| | Human subjects including the use of Embryonic Stem Cells |

Risk Assessment:

Please describe the risk assessment process and how the appropriate biosafety precautions were determined for this protocol; you may include database searched, search terms, dates, etc.

| Briefly describe Risk Assessment process and check off any database searches below: | |
|---|---|
| | |
| | |
| | PubMed Search |
| | CDC-NIH Guidelines |
| | rDNA Guidelines |
| | Stevens Institute Safety Literature |
| | Other: |

Protocol Specific Laboratory Safety:
Please complete Section G for all protocols submitted to the Biosafety Committee for consideration.

**Principal Investigator Acknowledgement:**

By signing below, the Principal Investigator acknowledges that the laboratory workers (including students, faculty, staff or visitors) under his or her direction have received appropriate training required to manipulate, store, and disinfect the microorganisms, human-derived materials, or recombinant materials proposed for use in the following protocol. Further, laboratory workers have been instructed on emergency procedures involving potentially infectious materials as outlined in Section VI (page 37) of the Stevens Biological Safety Guide.

Principal Investigator: _____ Date: _____

**Biosafety Committee Action**:

This protocol was reviewed by the Stevens Institutional Biosafety Committee on: _____

The following IBC action was taken:

| | |
|---|---|
| | Protocol Approved |
| | Protocol Withdrawn |
| | Protocol Tabled Until Next Meeting |
| | Protocol Not Approved |

Approved Protocols:

| | | | |
|---|---|---|---|
| | Assigned Biosafety Level: | | |
| | Protocol approved by: (sign and date below) | Signature | Date |
| | • Stevens Biosafety Officer | | |
| | • Chair, Stevens Biosafety Committee | | |
| | Protocol will remain in effect until | | |
| | • BL-1 Protocols are approved for 3 years | | |
| | • BL-2 Protocols are approved for 2 years | | |
| | Associated IACUC Protocol #: | | |
| | Associated IRB Protocol #: | | |

**Section B: <u>Exempt Recombinant DNA Experiments</u>** (please check those sections of the NIH Guidelines under which your experiments are exempt)

| | |
|---|---|
| | **Section III-F-1**. Those synthetic nucleic acids that: (1) can neither replicate nor generate nucleic acids that can replicate in any living cell (e.g., oligonucleotides or other synthetic nucleic acids that do not contain an origin of replication or contain elements known to interact with either DNA or RNA polymerase), and (2) are not designed to integrate into DNA, and (3) do not produce a toxin that is lethal for vertebrates at an LD50 of less than 100 nanograms per kilogram body weight. If a synthetic nucleic acid is deliberately transferred into one or more human research participants and meets the criteria of Section III-C, it is not exempt under this Section. |
| | **Section III-F-2**. Those that are not in organisms, cells, or viruses and that have not been modified or manipulated (e.g., encapsulated into synthetic or natural vehicles) to render them capable of penetrating cellular membranes. |
| | **Section III-F-3**. Those that consist solely of the exact recombinant or synthetic nucleic acid sequence from a single source that exists contemporaneously in nature. |
| | **Section III-F-4**. Those that consist entirely of nucleic acids from a prokaryotic host, including its indigenous plasmids or viruses when propagated only in that host (or a closely related strain of the same species), or when transferred to another |
| | **Section III-F-5**. Those that consist entirely of nucleic acids from a eukaryotic host including its chloroplasts, mitochondria, or plasmids (but excluding viruses) when propagated only in that host (or a closely related strain of the same species). |
| | **Section III-F-6**. Those that consist entirely of DNA segments from different species that exchange DNA by known physiological processes, though one or more of the segments may be a synthetic equivalent. A list of such exchangers will be prepared and periodically revised by the NIH Director with advice of the RAC after appropriate notice and opportunity for public comment (see Section IV-C-1-b-(1)-(c), Major Actions). See Appendices A-I through A-VI, Exemptions under Section III-F-6--Sublists of Natural Exchangers, for a list of natural exchangers that are exempt from the NIH Guidelines. |
| | **Section III-F-7**. Those genomic DNA molecules that have acquired a transposable element, provided the transposable element does not contain any recombinant and/or synthetic DNA. |
| | **Section III-F-8**. Those that do not present a significant risk to health or the environment (see Section IV-C-1-b-(1)-(c), Major Actions), as determined by the NIH Director, with the advice of the RAC, and following appropriate notice and opportunity for public comment. |
| | **Appendix C-VII**. The Purchase or Transfer of Transgenic Rodents |
| | **Appendix C-VIII**. Generation of BL1 Transgenic Rodents via Breeding<br>The breeding of two different transgenic rodents or the breeding of a transgenic rodent and a non-transgenic rodent with the intent of creating a new strain of transgenic rodent that can be housed at BL1 containment will be exempt from the NIH Guidelines if:<br>(1) both parental rodents can be housed under BL1 containment; and<br>(2) neither parental transgenic rodent contains the following genetic modifications: (i) incorporation of more than one-half of the genome of an exogenous eukaryotic virus from a single family of viruses; or (ii) incorporation of a transgene that is under the control of a gammaretroviral long terminal repeat (LTR); and<br>(3) the transgenic rodent that results from this breeding is not expected to contain more than one-half of an exogenous viral genome from a single family of viruses. |

Most experiments involving E. coli K-12 host vector systems and *Saccharomyces cerevisiae* and *Saccharomyces uvarum* host vector systems are exempt from the NIH Guidelines. If the answer to all 3 of the following questions are no, then the experiments are exempt according to Appendix C-II (for *E. coli* K-12) or Appendix C-III (for *Saccharomyces cerevisiae and Saccharomyces uvarum*).

| Yes | No | |
|---|---|---|
| | | Do any experiments involve Risk Groups 3, 4 or restricted organisms or nucleic acids from Risk Groups 3, 4 or restricted organisms? |
| | | Do any experiments involve introduction of genes coding for molecules toxic for vertebrates? |
| | | Will there be any large-scale experiments (more than 10 liters of culture)? |

**Section B: <u>Exempt Recombinant DNA Experiments</u> (Continued)**

Please include only information regarding **Exempt** rDNA experiments in the tables below.

| | **Host (s)** Indicate the host(s) into which the recombinant material (rDNA, RNA, virus) will be introduced. Examples incluse: *E. coli*, *S. cerevisiae*, human/animal cells, whole animals, plants. | **Species** Subspecies, variety, serotype, strain. | **Vectors** Which host-vector system will be used for this research? Examples include: bacterial plasmids, yeast plasmids, cultured cell plasmid vectors, baclovirus, AAV, other viral vectors | **DNA Sequence** List names of genes or DNA segments that will be evaluated | **Proteins** List proteins produced if applicable |
|---|---|---|---|---|---|
| #1 | | | | | |
| #2 | | | | | |
| #3 | | | | | |
| #4 | | | | | |
| #5 | | | | | |
| #6 | | | | | |

| Yes | No | |
|---|---|---|
| | | Will an attempt be made to purify any of the foreign gene products encoded by the gene? |
| | | Will a virus-derived vector system that is engineered to be replication-incompetent be used? |

## Section C: Non-Exempt Recombinant DNA Experiments

This section describes experiments covered by the NIH Guidelines. Check the appropriate registration category(s) for your experiment.

**Experiments that require IBC approval BEFORE initiation:**

| | |
|---|---|
| | **Section III-D-1-a**. Introduction of recombinant or synthetic nucleic acid molecules into risk group 2 agents |
| | **Section III-D-2-a**. Introduction of DNA from risk group 2 (or 3) agents into non-pathogenic bacteria or lower eukaryotes |
| | **Section III-D-3-a**. Use of infectious risk group 2 virus (or defective virus plus helper virus) in tissue culture systems |
| | **Section III-D-3-e**. Use of infectious risk group 1 virus (or defective virus plus helper virus) in tissue culture systems |
| | **Section III-D-4-a**. Transfer of recombinant or synthetic nucleic acid molecules EXCEPT for >2/3 of eukaryotic viral genomes into any non-human vertebrate or invertebrate organism |
| | **Section III-D-4-b**. Transfer of recombinant or synthetic nucleic acid molecules from risk group 2 (or higher risk group) human or animal pathogens into whole animals |

**Experiments that require IBC notification CONCURRENT WITH initiation:**

| | |
|---|---|
| | **Section III-E-1**. Experiments involving the formation of recombinant or synthetic nucleic acid molecules containing no more than 2/3 of the genome of any eukaryotic virus |
| | **Section III-E-2**. All components derived from non-pathogenic prokaryotes and non-pathogenic lower eukaryotes |
| | **Section III-E-3**. Experiments involving transgenic rodents |

**Some experiments require additional review/approval by NIH OBA before initiation:**

| | |
|---|---|
| | **Section III-A-1-a**. Transfer of a drug resistant gene into microorganisms that do not acquire the gene naturally that could compromise use of the drug to control disease in humans, veterinary medicine or agriculture |
| | **Section III-B-1**. Cloning of genes for toxins with LD50 of > 10 ng/kg body weight |

If your non-exempt research does not fall into any of the categories listed above, review Section III of the NIH Guidelines and use the space below to provide a brief description of the research and the appropriate NIH Guidelines referenced.

Section of the NIH Guidelines:

Description:

## Section C: Non-Exempt Recombinant DNA Experiments (Continued)

Generation and Use of rDNA

Complete this section if you are generating and/or using non-exempt rDNA in your laboratory.

Answer questions 1-8 for EACH host-vector system.

Transgene

1. Describe the gene sequence(s) inserted into the recombinant vector:

   a. Source of gene(s) (genus/species):

   b. Do any of the gene sequences increase oncogenic potential, originate from an HHS or USDA select agent or toxin, transfer a drug resistance trait that has the potential to compromise the use of the drug to control disease or have the potential to increase the pathogenicity or virulence of a vector system?
   No

   Yes, explain below:

   c. Describe the function and activity of the transgene(s):

   If you are planning on using an extensive number of transgenes, list classes.

   If you are using a genome-wide approach, indicate the components of the constructs in the library or libraries.

2. If any of the above genes are from a viral source, do they compromise more than 2/3 of the viral genome?
   No

   Yes, specify.

3. Will a deliberate attempt be made to obtain expression of the foreign gene encoded in the recombinant DNA or RNA?
   No

   Yes

4. Identify vector system - Please only describe one host-vector system:

| | |
|---|---|
| | Bacterial Plasmid |
| | Adeno-Associated Virus |
| | Adenovirus |
| | Simple Retrovirus |
| | Lentivirus |
| | Viruses other than lentivirus, simple retrovirus, adenovirus or adeno-associated virus |
| | Describe: |

5. List host cell line or packaging cells for recombinant vector propagation:

6. Viral vector system(s)

   a. What % of the viral genome remains?

   b. Is a helper virus required for replication?

      No

      Yes

7. Target Recipient(s) - Indicate the recipient(s) of the DNA (check all that apply):

| | |
|---|---|
| | Bacterial Cells |
| | Animal Cells in Culture |
| | Animals |
| | Modified Tissue Culture Cell Lines into Animals |
| | Plant Cells |
| | Plants |
| | DNA Vaccine, specify target recipient(s) |

8. Investigators assessment of risk – This work will be conducted at (check appropriate biosafety level):

| | |
|---|---|
| | Biosafety Level 1 |
| | Biosafety Level 2 |

If you need to describe additional non-exempt host-vector systems, please make a copy of pages 6 and 7 and answer questions 1 – 8 for each host non-exempt host vector system.

**Section D: <u>Research with Potentially Infectious Biological Agents</u>**

Complete this section if you are working with an agent that could cause an infection in humans. Provide the information requested below for each agent.

| | Yes | No | Please Provide Details Below |
|---|---|---|---|
| 1. Name of agent (include genus, species, sub-species, strain, etc.): | | | |
| 2. Will antibiotic resistance be expressed? | | | |
| 3. Will toxin be produced? | | | |
| 4. Largest volume of agent to be cultured? | | | |
| 5. Will agent be concentrated? | | | |
| 6. If agent is to be concentrated, how will it be concentrated? | | | |
| 7. How frequently will agent be manipulated? | | | |
| 8. How will agent be inactivated? | | | |
| a. heat | | | |
| b. chemical | | | |
| c. other (list): | | | |
| 9. Will agent be introduced into animals? | | | |
| 10. Have all personnel that will be handling this agent received appropriate biosafety training? | | | |
| | | | |
| If you need to describe additional potentially infectious agents, please make a copy of page 8 and answer questions 1 – 10 for each potentially infectious agent. | | | |

**Section E: Human and Non-human Primate Blood, Body Fluids and Cell Lines**

Identify the type and source of the materials to be used:

1. Samples to be manipulated (for human or non-human primate cells lines, indicate if cells are established or primary):

2. Source of samples:

3. If commercially obtained, please list vendor and specific cell lines:

4. Have all personnel who work with human material completed the appropriate bloodborne pathogens training program?

5. Is laboratory equipped with biological safety cabinet or other containment equipment to safely manipulate these materials?

If you need to describe additional materials or cell lines, please make a copy of page 9 and answer questions 1 – 4 for each cell line.

**Section F: <u>Toxins of Biological Origin</u>**

Complete this section if you are working with a toxin of biological origin. Provide the information requested below for each toxin.

1. Name of toxin:

2. Largest quantity in use and stored:

3. Describe how the toxin is stored:

4. Describe the toxin deactivation/disposal procedures:

If you need to describe additional toxins, please make a copy of page 10 and answer questions 1 – 4 for each toxin.

Section G: Protocol Specific Laboratory Safety

1. Personnel and Training

Please list all laboratory personnel involved in this protocol and indicate the dates of the required training. If training has not yet been scheduled, please indicate pending or TBD.

| Name | Title | Date of Biosafety Training | Date of BBP Training | Other Protocol Specific Training |
|---|---|---|---|---|
| | | | | |
| | | | | |
| | | | | |
| | | | | |
| | | | | |

2. Containment Equipment

Please list type and location of containment equipment (e.g., biological safety cabinet) and date of certification if required.

| Containment Equipment | Type | Certification Date | |
|---|---|---|---|
| Biological Safety Cabinet | | | |
| Centrifuge with Safety Caps and Sealed Rotors | | | |
| Splash Guard | | | |
| Other: | | | |
| | | | |

3. Equipment and Surface Decontamination

Please list the decontamination solution used, concentration, and frequency for various laboratory equipment and work surfaces.

| Equipment and/or Work Surfaces | Decontamination Solution | Concentration | Frequency |
|---|---|---|---|
| Biological Safety Cabinet | | | |
| Laboratory Bench | | | |
| Mechanical pipetter | | | |
| | | | |
| | | | |

4. Spill Control

Please describe available laboratory spill control equipment and procedures.

5. Waste Decontamination

Please describe how potentially contaminated laboratory waste is decontaminated and subsequently disposed.