University of Tennessee

User's Guide for the Institutional Biosafety Committee Registration Form

A reference for researchers of UT Knoxville, UT Institute of Agriculture, and the Graduate School of Medicine for completing and understanding the Institutional Biosafety Committee registration process

Table of Contents

Introduction

This document has been developed to assist researchers in preparing registration documents for review by the University of Tennessee Institutional Biosafety Committee (IBC). The IBC registration form is designed to capture information about facilities, personnel, and research projects involving the following items:

- Recombinant DNA (rDNA)
- Infectious Agents
- Human Derived Materials
- Acute Biological Toxins (LD^{50} < 100 ng/kg) & Select Agent Toxins
- Venomous Animals
- Poisonous Plants

Recombinant DNA (rDNA)

In the context of the *NIH Guidelines*, recombinant DNA molecules are defined as either: (i) 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 (ii) molecules that result from the replication of those described in (i) above.

Synthetic DNA segments which are likely to yield a potentially harmful polynucleotide or polypeptide (e.g., a toxin or a pharmacologically active agent) are considered as equivalent to their natural DNA counterpart. If the synthetic DNA segment is not expressed in vivo as a biologically active polynucleotide or polypeptide product, it is exempt from the *NIH Guidelines*.

Genomic DNA of plants and bacteria that have acquired a transposable element, even if the latter was donated from a recombinant vector no longer present, are not subject to the *NIH Guidelines* unless the transposon itself contains recombinant DNA.

Infectious Agents

For purposes of this registration the following types of infectious agent research must be registered with IBC:

- Microorganisms capable of causing disease in healthy human adults (Risk Group 2 or higher)
- Animal pathogens requiring enhanced containment per USDA APHIS regulatory permit specifications
- Plant pathogens requiring enhanced containment per USDA APHIS regulatory permit specifications
- Non-human primate cells/cell lines (all human cell lines must be listed in the Human-Derived Materials Section, see below).
- Field procedures with a high risk for infectious disease transmission.

Human Derived Materials

Completion of this section is required if your research includes the use of human blood, blood products, trauma fluids, unfixed tissues, or human cell lines or cultures.

Acute Biological Toxins (LD^{50} < 100 ng/kg) & Select Agent Toxins

Infectious agents and toxins that are considered by the Department of Health & Human Services (DHHS) or the United States Department of Agriculture (USDA) as having the potential to pose substantial harm or a severe threat to human, animal or plant health or plant products are regulated as "select agents."

For more information on select agents and toxins, refer to the Biosafety Office website at: http://biosafety.utk.edu/agents/

IBC Registration Process

To register your research with the Institutional Biosafety Committee, you must complete a registration document and submit it to:

UT Biosafety Office
336 Ellington Plant Sciences Bldg.
2431 Joe Johnson Drive
Knoxville, TN 37996-4564

A copy of the completed form can be submitted by clicking on the "Submit by E-mail" button at the very end of the form or via email to branger@utk.edu to initiate the review process. Final IBC approval of the registration will not be granted until a hard copy signed by the Principal Investigator (PI) and appropriate Department Head is received by the Biosafety Office and approved by the IBC Chair.

Click here to access the "IBC Registration" form or visit the UT Biosafety Office website (http://biosafety.utk.edu) to access this form under the "Forms" link on the main page.

The "Form Preparation Pointers" section of this document (p. 4) will provide you with guidance for form completion to ensure that all necessary information is completed appropriately to minimize the potential for delays in the IBC review process. A sample of a completed form is included for your reference as well.

Incomplete forms will not be sent on to the IBC for review, but will be returned to the submitting Principal Investigator (PI). The IBC meets on a monthly basis (typically the third Wednesday of the month) to review and vote on registrations submitted during the previous calendar month. Therefore, if you want to have your registration on the month's agenda, you should submit a completed registration document to the Biosafety Office no later than the **first Friday of the month** so that all IBC deadlines can be met.

If your work will require biosafety level 2 (BSL-2) practices and containment, or your work falls into a Category III-A, III-B, III-C, or III-D of the *NIH Guidelines*, it is strongly recommended that you contact the Biosafety Office for assistance with form preparation at (865) 974-1938.

You should attend the IBC meeting when your proposed work will be reviewed if at all possible. Your presence at the meeting will greatly facilitate the communication process. If you do not attend, and questions arise that cannot be addressed, you will most likely receive a request for more information and further review of your proposed protocol will be delayed.

Once approval is granted by the IBC, an approval letter and a copy of the approved version of the registration document will be generated by the Biosafety Office. Whenever possible, approval letters will be prepared and sent to PI's within the same business month. Contact the Biosafety Office if you do not receive any communication regarding the outcome of the review process by the end of the month for which your review was scheduled.

IBC approval of protocols will be valid for 3 years from the approval date. However, in order to maintain your approval, you must complete and submit an annual updates to document any changes in the research materials or techniques listed on the original registration. You will receive a notice from the Biosafety Office approximately one month before the anniversary date of your approval with instructions for form completion and submission. Contact the Biosafety Office if you do not receive such a notice before your approval anniversary date.

Note that if your recombinant DNA work changes to the degree where the work falls into a different review category, or presents new considerations for containment requirements, you must notify the IBC Chair before initiating these changes to determine if further review actions are required.

It is your responsibility and in your best interest to take measures to ensure that your work is properly documented and currently approved to assure compliance with regulations and policies that can impact funding.

Form Preparation Pointers

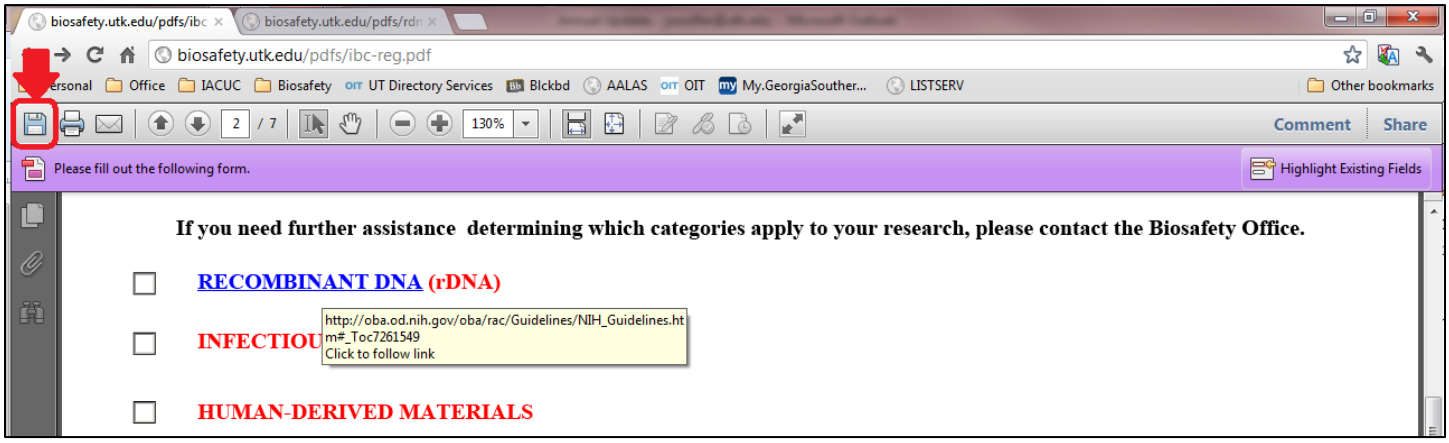
In order for you to complete this form electronically, it will be necessary to download an updated version of Adobe Acrobat Reader (preferably 9 or higher). A free version can be obtained at: http://get.adobe.com/reader/. (Contact OIT at (865) 974-9900 for additional help with installing or updating your version of Adobe Acrobat Reader).

The IBC registration form is divided into seven sections as follows:

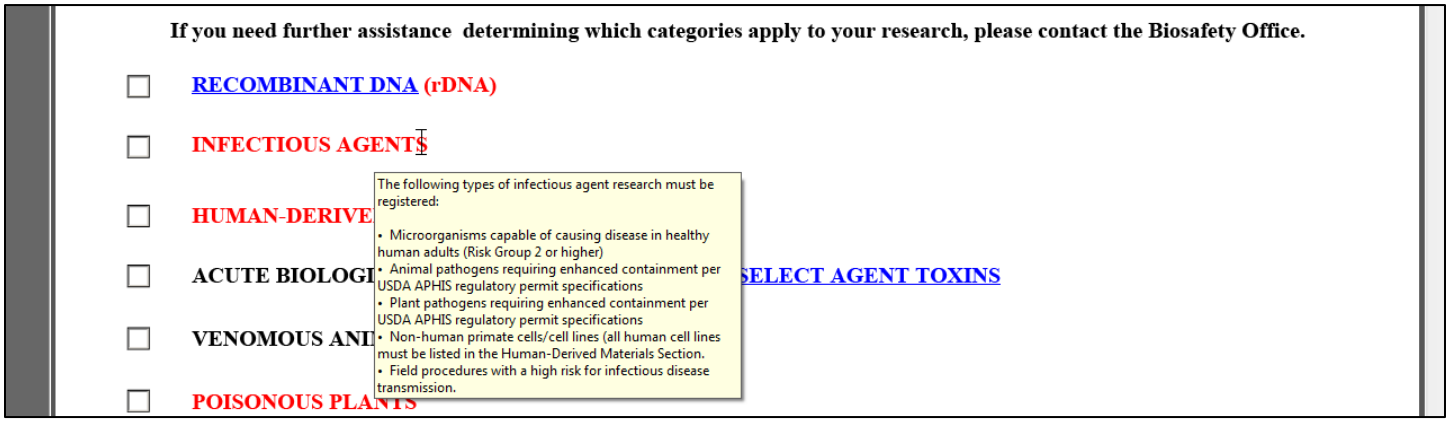
- Section I: General Information
- Section II: Non-Technical Summary
- Section III: Type of Research
- Section IV: Technical Summary
- Section V: Facilities & Procedures Assessment
- Section VI: Personnel
- Section VII: Acknowledgement and Approval

In each section there are items with color coded text to bring additional information about completing the form to your attention. All seven sections are color coded as **ORANGE** and are required to be completed in their entirety. However, Section III: Type of Research only requires you to select the appropriate categories for your research registration and complete the sub-sections within those categories that follow.

Items with text color coded in **BLUE** and **UNDERLINED** have hyperlinks that will take you to an external website for additional information on that item. ***(Note: you should save the form before selecting a hyperlink to prevent accidental loss of entered information).***



Items with text in **RED** have additional information that can be viewed by scrolling over the text.



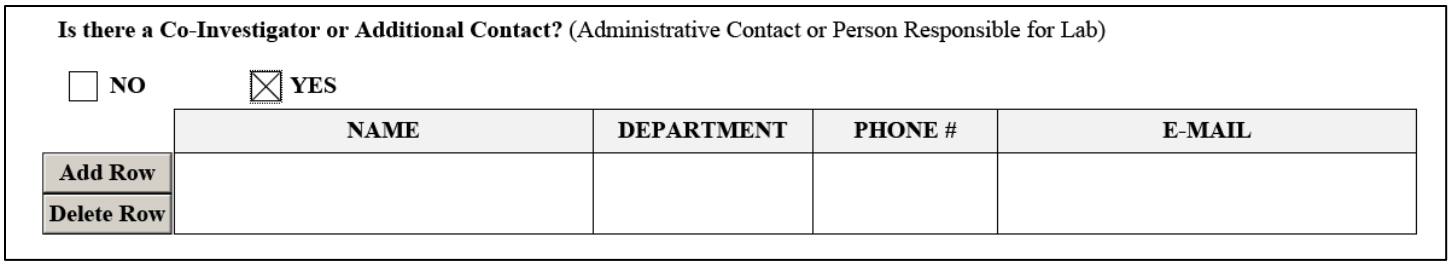
Section I. General Information

Principal Investigator

The *NIH Guidelines* refer to the researcher who is responsible for documentation, training and reporting relative to the recombinant DNA molecule use as the "Principal Investigator". For registration with the UT IBC, the Principal Investigator is someone who has a faculty appointment with UT and meets funding agency requirements as a lead researcher. If you do not have such an appointment, you must partner with someone who does and work with that individual to have the work appropriately documented through that individual on your behalf.

Under this section enter the full name of the principal investigator along with job title, department, mailing address, contact phone number, and email address.

If there are additional administrative contacts or lab personnel responsible for this registration, check "YES" and complete the table with the contact information for the additional contacts by providing their name, department, phone number, and email address. To add more than one contact, click the "Add Row" button located to the left of the table.



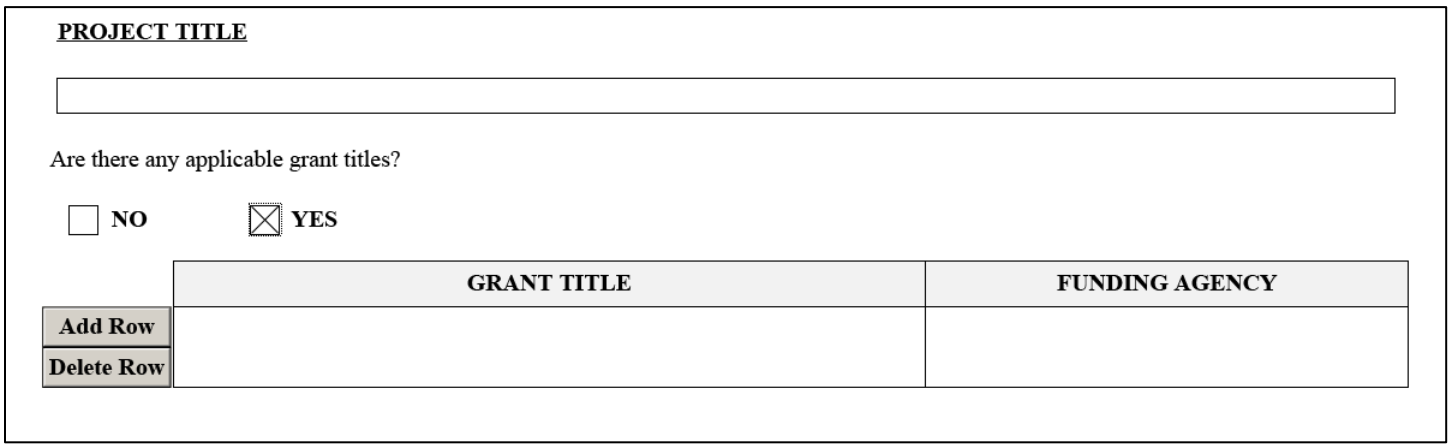
Is there a Co-Investigator or Additional Contact? (Administrative Contact or Person Responsible for Lab)

☐ NO ☒ YES

	NAME	DEPARTMENT	PHONE #	E-MAIL
Add Row				
Delete Row				

Project Title

This section is expandable and the title should represent the research captured in the registration. It should not be specific for a particular grant proposal if the registration is intended to cover multiple grant proposals. IBC approval letters will list the general project title. If you find that you need an approval letter that specifies a particular grant title, you can request such a letter from the Biosafety Office if this grant title is listed on the approved registration. To list grant titles and funding agencies applicable to this registration, click "Yes" under the question "Are there any applicable grant titles?" To add additional rows, click the "Add Row" button to the left of the table.



PROJECT TITLE

Are there any applicable grant titles?

☐ NO ☒ YES

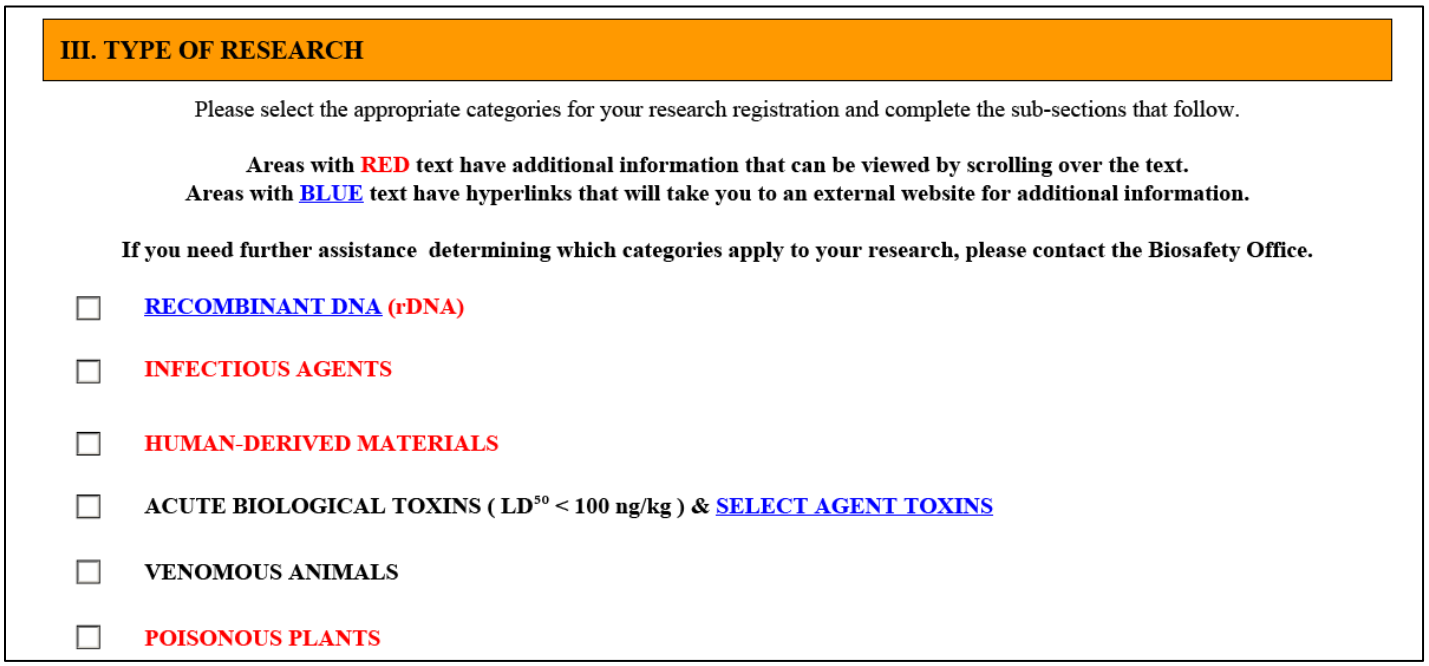
	GRANT TITLE	FUNDING AGENCY
Add Row		
Delete Row		

Section II. Non-Technical Summary

This section is expandable but is limited to 4,000 text characters (roughly half a page). The response to this question will help to clarify the information provided within this registration in a manner that can be easily understood by reviewers who are not experts in your field including regulatory auditors. The non-technical summary should capture the general aims and significance of your research. Refrain from using technical jargon and/or giving detailed procedures.

Section III. Type of Research

This section lists the categories of research under the purview of the IBC. Select all categories applicable to your research. Once the boxes are checked, sub-sections will automatically appear based on the selection(s) made.



Recombinant DNA (rDNA)

Question 1

Mark each box in A-I that pertains to your registered rDNA research:

A. *Deliberate transfer of drug-resistance traits to microorganisms of clinical, environmental or agricultural significance that could compromise the use of drugs to control disease.* This does not refer to antibiotic-resistance selectable markers of no clinical significance or those used as screening tools in low-risk organisms.
B. *Clinical trial of human gene therapies.* This item refers to any work involving human gene transfer protocols/clinical trials (Category III-C of the *NIH Guidelines*).
C. *Work with a Risk Group 2 or higher pathogen.* This applies to any microorganisms that are infectious to immunocompetent humans and may apply to certain animal or plant pathogens per USDA APHIS (Animal & Plant Health Inspection Service) containment requirements.
D. *Work with a Select Agent or high-consequence pathogen.* This item has links to the DHHS and USDA Select Agents and Toxins list as well as the *NIH Guidelines*, Appendix B describing the basis for classification of Biohazardous Agents by Risk Group.
E. *Work with live animals.* This item populates a sub-section wherein IACUC protocol approval numbers are to be entered as well as information regarding rDNA or rDNA-modified organisms to be used in the animals. Click either the "Yes" or "No" boxes provided in this section as applicable. If "Yes" for E-1, describe how DNA will be introduced and the source in the expandable text boxes provided in E-2 & E-3 respectively. If you selected "Yes" for questions E-4 to E-5, complete the expandable text boxes that appear by providing a detailed description for each question.

E. ☒ Work with live animals (i.e., genetic modification of animals or challenge of animals with recombinant DNA or viable recombinant DNA-modified organisms).

IACUC #: _____

1. Does this study involve the alteration of the germ line of the animals? ☐ **YES** ☐ **NO**

2. How will DNA be introduced?

3. What is the source of the DNA?

4. Will rDNA modified organisms be used in animals? ☒ **YES** ☐ **NO**

List organisms to be used:

5. Does this experiment use viruses as host-vector systems? ☒ **YES** ☐ **NO**

a. Do the experiments involve formation of rDNA molecules containing > two-thirds of the genome of any eukaryotic virus? ☒ **YES** ☐ **NO**

Please describe:

b. Do the experiments involve the use of infectious human or animal viruses? ☒ **YES** ☐ **NO**

Please describe:

c. Do the experiments involve the use of a defective human or animal virus in the presence of a helper virus? ☒ **YES** ☐ **NO**

Please describe:

F. Work with live plants. This refers to genetic modification of plants or challenge of plants with recombinant DNA-modified organisms to include microbes and arthropods. A series of "Yes" or "No" questions help further evaluate and assess the necessary containment or safety procedures required for projects involving live plants or associated organisms. Click either "Yes" or "No" as they apply to the genetically modified plants/organisms to be used. A table is also provided to list all transgenic or genetically modified whole plants. To add additional rows click the "Add Row" button to the left of the table.

F. ☒ Work with live plants (i.e., genetic modification of plants or challenge of plants with recombinant DNA-modified organisms to include microbes and arthropods).

YES	NO	
☐	☐	The production of sexual or asexual botanical reproductive structures of transgenic plants that have the potential for being released.
☐	☐	Plants modified by rDNA that are noxious weeds or that can interbreed with noxious weeds.
☐	☐	Plants in which the introduced DNA represents the complete genome of an infectious agent.
☐	☐	Plants associated with rDNA-modified microorganisms that **DO** have a recognized potential for serious detrimental impact on managed or natural ecosystems.
☐	☐	Plants associated with rDNA-modified microorganisms that **DO NOT** have a recognized potential for serious detrimental impact on managed or natural ecosystems.
☐	☐	Experiments with rDNA-modified arthropods or small animals associated with plants, or with arthropods or small animals with recombinant DNA-modified microorganisms associated with them if the recombinant DNA-modifed microorganisms **DO** have a recognized potential for serious detrimental impact on managed or natural ecosystems.
☐	☐	Experiments with rDNA-modified arthropods or small animals associated with plants, or with arthropods or small animals with recombinant DNA-modified microorganisms associated with them if the recombinant DNA-modifed microorganisms **DO NOT** have a recognized potential for serious detrimental impact on managed or natural ecosystems.

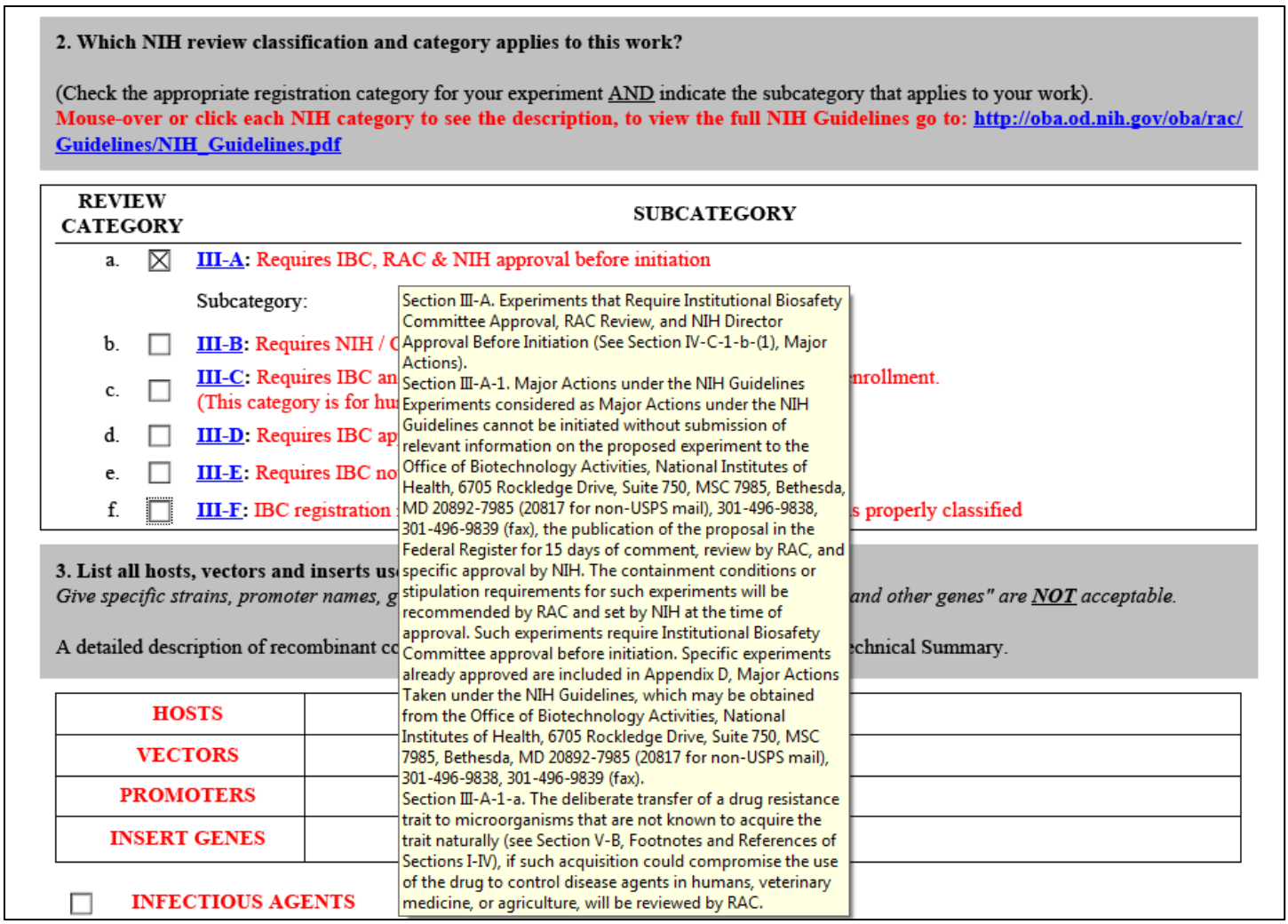
Please list ALL transgenic / genetically modified whole plants:

	PLANT (GENUS AND SPECIES)	INSERTED DNA & FUNCTION DESCRIPTION	BIOLOGICAL SOURCE OF INSERTED DNA
Add Row			
Delete Row			

G. Work involving cloning of genes that encode for toxins. This box should only be checked if cloning toxins that have the potential to affect mammalian health, particularly toxins with an $LD^{50} < 100$ ng/kg as determined in mammalian species.
H. Work that will involve more than 10 liters of culture in one vessel.
I. Not applicable. Check this item if none of the above items pertain your registration.

Question 2

To determine your review category and subcategory, scroll over the text in **RED** to view a brief description of each NIH category. To view the full NIH Guidelines go to: http://oba.od.nih.gov/oba/rac/Guidelines/NIH_Guidelines.pdf. Click the review category that applies to your work. A "Subcategory" box will appear in which you will enter the specific NIH subcategory listed in the NIH Guidelines. (For example, the illustration below displays how the form would be completed if your work involves the deliberate transfer of drug resistant trait to a microorganism not known to acquire this trait naturally and which compromises disease control in medicine, agriculture, etc.).



Question 3

This table is expandable and is intended to capture all hosts, vectors and inserts used as part of your work. You should be as specific as possible with listing the materials that you may use. Vectors, promoters, and insert genes may be grouped by type/function provided that they are adequately described and/or examples are provided. It is understood that this list is likely to change through the research process. For this reason, you will be expected to complete an annual update to document any changes in your rDNA procedures as described in the "IBC Review Process" section of this guide.

NOTE: *If your approved recombinant DNA research will change to the degree that it alters the NIH review category or presents new considerations for containment requirements, you must notify the IBC Chair before implementing the changes to determine if further IBC review is required.*

- *Host* examples include bacteria, yeast, cell lines, plants, and animals. Include species, strain, or technical names in this section.
- *Vectors* used for rDNA delivery are to be listed by technical name whenever possible; if not, they may be listed by type or function (e.g. shuttle, expression, lentiviral or retroviral, transposon, etc.).
- Listed *promoters* should be described by source (organism of origin) and activity (e.g. inducible, constitutive, etc.).
- *Insert genes* may be identified by gene/gene family name or protein(s) encoded. Whenever possible, details such as the natural source, activity, and resultant phenotype should be given (may be included in Section IV, Technical Description) as this information will help the committee conduct a thorough risk assessment of the recombinant constructs and hosts. All-inclusive or vague terms like "other genes", "similar genes", or "associated genes" are generally not acceptable.

3. List all hosts, vectors and inserts used as part of this protocol in the table below.
*Give specific strains, promoter names, gene families, or class at a minimum. Catchalls like "..and other genes" are **NOT** acceptable.*

A detailed description of recombinant construct development must be given in Section IV - Technical Summary.

HOSTS	
VECTORS	
PROMOTERS	
INSERT GENES	

Infectious Agents

Question 1

This table is expandable and is intended to capture information infectious agents that will be used in your research projects. The following must be listed under the first column as the scientific name of the agent or description of the materials:

- Microorganisms capable of causing disease in health human adults (Risk Group 2 or higher)
- Animal pathogens requiring enhanced containment per USDA APHIS regulatory permit specifications
- Plant pathogens requiring enhanced containment per USDA APHIS regulatory permit specifications
- Non-human primate cells/cell lines (all human cell lines must be listed in the Human-Derived Materials Section)
- Field procedures with a high risk for infectious disease transmission

In the second column, click the box for the appropriate Risk Group that the infectious agent would fall under. According to the U.S. Department of Health and Human Services publication of the 5th Edition of *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, Risk Groups are classified as follows:

- RG-1 (Risk Group 1): Agents that are not associated with disease in healthy adult humans
- RG-2 (Risk Group 2): Agents that are associated with human disease which is rarely serious and for which preventive or therapeutic interventions are *often* available
- RG-3 (Risk Group 3): Agents that are associated with serious or lethal human disease for which preventive or therapeutic interventions *may* be available (high individual risk but low community risk)
- RG-4 (Risk Group 4): Agents that are likely to cause serious or lethal human disease for which preventive or therapeutic interventions are *not usually* available (high individual risk and high community risk)

In the third column list any features or characteristics of the infectious agent that could impact containment practices such as antimicrobial resistance, production of toxins, attenuation, etc.

In the fourth column click the appropriate box to indicate the origin or source of the infectious agent from the options provided. If you select “Other”, specify in the text box below the row the origin or source of the infectious agent.

1. List the Infectious Agents to be used

The following must be listed:

- Microorganisms capable of causing disease in healthy human adults (Risk Group 2 or higher)
- Animal pathogens requiring enhanced containment per USDA APHIS regulatory permit specifications
- Plant pathogens requiring enhanced containment per USDA APHIS regulatory permit specifications
- Non-human primate cells/cell lines (all human cell lines must be listed in the Human-Derived Materials Section.
- Field procedures with a high risk for infectious disease transmission.

	SCIENTIFIC NAME OF AGENT OR DESCRIPTION OF MATERIALS	RISK GROUP (RG) ASSIGNED	CHARACTERISTICS (List any features that could impact containment practices such as antimicrobial resistance, production of toxins, attenuation, etc.)	ORIGIN / SOURCE
Add Row Delete Row		☐ RG-1 ☐ RG-2 ☐ RG-3 ☐ RG-4		☐ Directly from human/animal/ plant tissue ☐ On-campus Collection / Researcher ☐ Off-campus Collection ☐ Off-campus Researcher ☐ Laboratory Stock ☒ Other

Please Specify:

Question 2

This table lists a series of "Yes" or "No" questions to help further evaluate and assess the necessary containment or safety procedures required for projects involving infectious agents. Click "Yes" or "No" as appropriate to the procedures or processes that will be used in your research project. Questions that are answered as "Yes" on Rows 1 and 4 will have sub fields appear requiring additional clarification or information as shown below.

2. Please mark "YES" or "NO" for each question.

PROJECT ASSESSMENT

YES	NO	
☒	☐	Processes involving concentration of infectious agents.
		☐ Subculturing or Propagation ☐ Filtration / Sedimentation & Small-Volume Resuspension ☐ Centrifugation & Small-Volume Resuspension ☒ Other
Please Explain:		
☐	☐	Generating of cultures containing infectious agent(s) in quantities of greater than 1 liter
☐	☐	Use of sharps in conjunction with an infectious agent(s)
☒	☐	Use of animals in conjunction with an infectious agent(s)
		IACUC #:
☐	☐	Use of recombinant DNA molecules in conjunction with an infectious agent(s)
☐	☐	Procedures with a high potential for aerosol generation (i.e., centrifugation, vortexing, shaking, etc.) in conjunction with an infectious agent

Question 3

The final question under the Infectious Agents section lists a series of questions associated with field procedures that may be used in conjunction with your research project. If you will not be conducting any of the listed procedures, click in the "N/A" box and the list of procedures will be hidden. If you place choose any of the procedures boxes listed, you must give a description of the field procedures and applicable safety precautions in Section IV, Technical Summary.

3. High Risk Field Procedures: Check all that apply. A description of field procedures and associated experiments must be given in Section IV. Technical Summary. If you are not conducting any of the following procedures, click N/A. ☐ N/A

- ☐ Trapping and handling of wild animals for epidemiological surveillance of agents infectious to humans designated as Risk Group 2 or higher;
- ☐ Trapping and handling of wild animals that may transmit significant or life threatening zoonotic diseases through bites, scratches, or aerosols (e.g. rabies, hantavirus, tuberculosis, etc.) as determined by risk assessment of the target species and proposed procedures;
- ☐ Collecting arthropod (or other) vectors of human disease for purposes of epidemiological surveillance (e.g. mosquito collection for West Nile virus testing; tick collection for Lyme disease testing);
- ☐ Collecting environmental samples that have a high risk of containing significant or life threatening infectious agents as determined by risk assessment of the samples and proposed procedures;
- ☐ Isolating microorganisms from any of the above for purposes other than routine identification and enumeration;

Human-Derived Materials

Question 1

The following table is expandable and to add rows click the "Add Row" button or to delete rows click on the "Delete Row" button. Under the first column list types of human-derived materials such as blood, body fluids, or cell lines. (If listing a cell line, provide the cell line name, such as HeLa cells).

In the second column give a brief description of the material and include the tissue origin of cell lines (e.g., cervical carcinoma).

In the third column provide the source or vendor of the human-derived materials. If derived from a patient, indicate "primary tissue" or "diagnostic specimen."

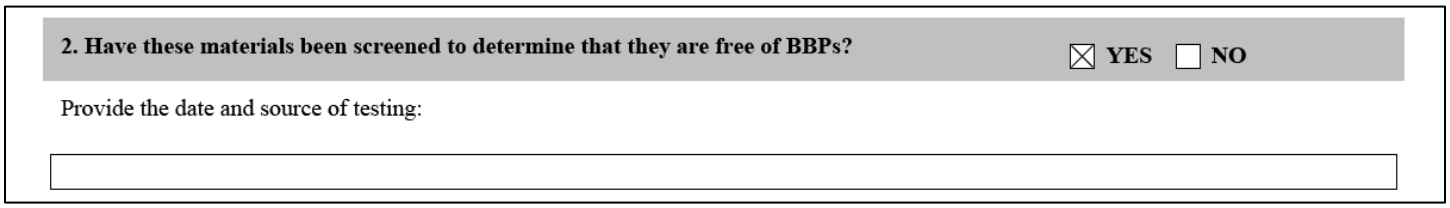
☒ **HUMAN-DERIVED MATERIALS**

1. Please complete the table below for all human-derived materials to be used:

	TYPE OF MATERIAL	BRIEF DESCRIPTION OF MATERIAL INCLUDING TISSUE ORIGIN OF CELL LINES	SOURCE / VENDOR OF MATERIAL
Add Row / Delete Row			

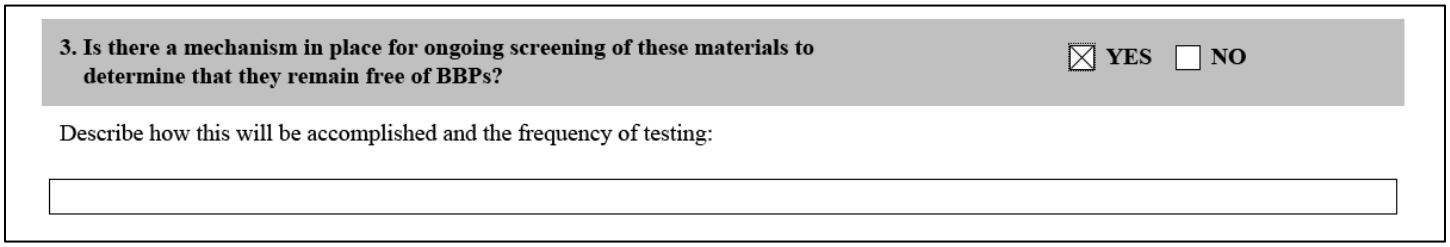
Question 2

The following question is used to determine if the human-derived materials have been screened for bloodborne pathogens. If they have been screened, click the "Yes" box and provide the date and source of testing the explanation box that appears. If they have not been screened, click in the "No" box and proceed to Question 3.



Question 3

This question is a follow up to Question 2 above. If materials are not prescreened to be free of BBPs, then "No" should be indicated by default. For those materials initially screened and confirmed to BBP free, you need to indicate whether there will be ongoing testing of the materials to assure that they remain BBP-free throughout your research. If there is a mechanism in place, click "Yes" and provide a description of how the screening is accomplished and the frequency in which the materials will be tested. If there is not a mechanism in place, indicate "No" box and proceed to the next applicable section.



Acute Biological Toxins (LD^{50} < 100 ng/kg) & Select Agent Toxins

The following section asks a series of eighteen expandable question boxes regarding toxins used in your research registration. Answer every question within this section. A copy of the Materials Data Sheet (MSDS) for the toxins will also need to be submitted with your registration. See the section on "Submitting the Registration" located on page 21 for further details.

Question 1

List the name of the toxin(s) and any common synonyms that may be used for the toxin(s).

Question 2

Give LD^{50} values and the test species. (e.g., rats, mice, etc.) Refer to the toxin Material Safety Data Sheet (MSDS) for LD^{50} information.

Question 3

Describe any possible routes of exposure to the toxin(s).

Question 4

List the target organs or systems affected by the toxin(s).

Question 5

Describe the signs and symptoms associated with exposure to the toxin(s) in humans.

Question 6

List any available antidotes for the toxin(s). If there are available antidotes, you must provide information on location, accessibility, and administration of each antidote.

Question 7

Indicate where the toxin will come from; i.e. commercial supplier (provide company name), collaborator (name/institution), or generated/purified onsite?

Question 8

Describe the quantity of the toxin(s) that you intend to obtain for your research project.

Question 9

Indicate whether you will be obtaining the toxin in a form that will minimize handling and risk for personnel involved in your research project by placing an "X" in either the "Yes" or "No" boxes. If you indicated "No," explain why these forms are not an option for your research project by completing the expandable text box that appears.

9. Will you be obtaining toxin in a form that will minimize handling and risk for personnel? (i.e., premeasured vials, toxin in solution, etc.)

☐ **YES** ☒ **NO**

If No, please explain why these forms are not an option for your use:

Question 10

Describe how the toxin(s) will be prepared or reconstituted.

Question 11

Explain the quantity of toxin(s) required for each experiment conducted; i.e. how much will be used at one time.

Question 12

Describe the maximum quantity of toxin(s) that you plan to have in stock at any given time. Note that you must not exceed the *de minimis* levels of select agent toxins as none of the UT-Knoxville area campuses have an approved Select Agents program. You must contact the Biosafety Office if future research will require the use of select agent toxins in excess of *de minimis* levels established by the Select Agent regulations.

Question 13

Describe methods to be used to inactivate the toxin(s). You must also cite references that validate the methods of inactivation indicated.

Question 14

Describe the security measures that will be taken to ensure that the toxin(s) are secure. Also, explain how inventory control of the toxin(s) will be achieved.

Question 15

Indicate whether or not you agree to perform all high risk operations involving greater than or equal to LD^{50} concentrations or volumes of toxin with a minimum of two knowledgeable individuals present by placing an "X" in either the "Yes" or "No" boxes.

Each individual present must be familiar with the applicable procedures, maintain visual contact with the other, and be ready to assist in the event of an accident. Other personnel will not be present in the toxin use area when these procedures are underway.

Question 16

Indicate whether or not you agree to perform surface decontamination on all exposed environmental surfaces with an effective disinfectant for a contact time of 15 minutes at the conclusion of procedures involving toxins and toxin solutions by placing an "X" in either the "Yes" or "No" boxes.

This decontamination step will be followed by a wipe down cleaning of the surfaces with a general lab cleaner or ethanol to remove caustic or corrosive residues.

Question 17

Indicate whether or not you agree to post signage indicating "Toxins in Use-Personnel Only" in the room where procedures will be performed by placing an "X" in either the "Yes" or "No" boxes.

Any special entry requirements are to be posted on the entrance(s) to the room or discussed with lab personnel prior to the initiation of toxin work. Traffic flow of personnel into the toxin use area will be minimized while toxin work is underway. Visitors or contractors will not be permitted in the toxin use area when toxins are in use. The storage device for toxins will be posted with a "restricted access" notice including the name and number of the lead researcher.

Question 18

Indicate whether or not you agree to place primary containers of toxins and toxin solutions in secondary containers for transport and storage by placing an "X" in either the "Yes" or "No" boxes.

All secondary containers will be non-breakable, leak-proof and equipped with a means of closure when feasible. In the case of liquids, absorbent materials will be placed in the secondary container for further spill prevention. The secondary container for pure or concentrated forms of the toxin will have a lid or other means of closure and be closed with a lid for transport from one location to another.

Venomous Animals

The following section contains a series of six expandable text boxes and questions pertaining to the research use of venomous animals. If you intend to use venomous animals for your research, indicate so by checking the "Yes" box provided and completing the following:

Question 1

Describe the venomous animal(s) that are being used for your research project. Provide the scientific species name for each venomous animal to be used.

Question 2

Describe how the animal(s) will be used for in your research project.

Question 3

Provide a risk assessment to the environment and personnel involved in the research. Your response should include the health impact of an accidental envenomation (signs, symptoms, and consequences), number of personnel at risk, and training of those handling the venomous animal(s).

Question 4

Describe procedures that will be used to minimize the potential for bites from the venomous animals used for your research project.

Question 5

Describe what actions will be taken in the event of a bite (or other mechanism of envenomation, e.g. sting). You must include emergency response procedures to include 1) basic first aid; 2) availability of antivenin (or equivalent); and/or 3) emergency facilities to be used or contacted.

Question 6

Summarize the proposed containment procedures to be used including the rationale for selection. You should describe caging, standard operating procedures, and/or other handling requirements that minimize or eliminate the risk of escape or accidents.

☒ **VENOMOUS ANIMALS**

Will you be using venomous animals for research? ☒ **YES** ☐ **NO**

Please describe the venomous animal(s) to be used for research:

Please describe the research activities that the animal(s) will be used for:

Please provide a risk assessment to the environment and personnel:

Describe procedures used to minimize the potential for a bite:

What actions will be taken in the event of a bite:

Summarize the proposed containment procedures to be used including rationale for selection:

Poisonous Plants

The following section contains a series of six expandable text boxes and questions pertaining to the research use of poisonous plants posing a risk to humans via dermatological contact, inhalation, accidental ingestion or other route of exposure. If you intend to use poisonous plants meeting these criteria for your research, indicate so clicking the "Yes" box provided and completing the following:

Question 1

Describe the type(s) of poisonous plant(s) that are being used for your research project. Provide the scientific species name for each poisonous plant to be used.

Question 2

Describe how the poisonous plant(s) will be used in your research project.

Question 3

Provide a risk assessment to the environment and personnel involved in the research. Your response should include the health impact of an accidental exposure to plant poisons/toxins (signs, symptoms, consequences), number of personnel at risk, and training of those handling the poisonous plants.

Question 4

Describe procedures and/or precautions that will be taken to minimize exposure to plant poisons/toxins.

Question 5

Describe what actions will be taken in the event of exposure to plant poison(s). You must include emergency response procedures to include 1) basic first aid; 2) availability of medication and/or 3) emergency facilities to be used or contacted.

Question 6

Summarize the proposed containment procedures to be used including the rationale for selection. You should describe housing, standard operating procedures, and/or other handling requirements that minimize or eliminate the risk of accidental exposures.

☒ **POISONOUS PLANTS**

Will you be using poisonous plants posing a risk to humans via dermatological contact, inhalation, or other route of exposure for research? ☒ **YES** ☐ **NO**

Please describe the type(s) of poisonous plant(s) to be used for research:

Please describe the research activities that the plant(s) will be used for:

Please provide a risk assessment to the environment and personnel handling the plant(s):

Describe procedures and/or precautions to be taken in order to minimize exposure to plant poison:

Actions to be taken in the event of exposure:

Summarize the proposed containment procedures to be used including rationale for selection:

Section IV. Technical Summary

This section is expandable and instructions for completion of this section are detailed on the form and outlined as follows:

- Provide a technical description of the proposed research for this registration.
- Address the purpose and aims of the project including a brief but complete description and the methods used to conduct this research.
- Define all acronyms and include sufficient detail for the committee to make a risk assessment of the project.
- Descriptions of recombinant DNA work must include information that supports the review category of this research as classified under the *NIH Guidelines* and must reflect the materials identified for Section III - Recombinant DNA Question 3 (Hosts, Vectors, Promoters, Insert Genes Table).

Note that proper completion of this section is the key to facilitating completion of the IBC review and approval process. This field is not to exceed 10,000 text characters (approximately 2 pages in length). Additional resources pertaining to the technical summary should be submitted as an attachment along with this registration. Refer to the "Submitting the Registration" section on page 21 for further instructions.

Section V. Facilities & Procedures Assessment

This section outlines a thorough review of safety and containment measures in place for the facilities where the research outlined in the registration is being performed.

Question 1

List all locations where the work described in the registration will take place. Also select the biosafety level that applies to your work. To add rows, click the "Add Row" button to the left of the table.

Biosafety levels describe the work practices, engineering controls, administrative policies, and facility design that are necessary to protect human health and ensure physical containment when handling infectious agents (or diagnostic/environmental samples that may contain them), genetically modified organisms/microorganisms, certain types of cells/cell cultures, or biological toxins. Thorough descriptions of biosafety levels are found in the *NIH Guidelines* (Appendices G, K, P, and Q for laboratories, large-scale production facilities, greenhouses, and animals respectively) and the 5th Edition of *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*. General descriptions are as follows:

- Biosafety Level 1 (BSL-1) is suitable for work involving well-characterized agents not known to consistently cause disease in immunocompetent adult humans, and present minimal potential hazard to laboratory personnel and the environment.
- Biosafety Level 2 (BSL-2) is suitable for work involving agents that pose moderate hazards to personnel and the environment.
- Animal Biosafety Level 1 is suitable for work in animals involving well-characterized agents that are not known to cause disease in immunocompetent adult humans, and present minimal potential hazard to personnel and the environment.
- Animal Biosafety Level 2 is suitable for work involving laboratory animals infected with agents associated with human disease and pose moderate hazards to personnel and the environment.
- Plant Biosafety Level 1 (BL1-P) is used to provide a low level of containment for experiments involving transgenic plants in which there is no evidence that the modified organism would be able to survive and spread in the environment and, if accidently release, would not pose an environmental risk.
- Plant Biosafety Level 2 (BL2-P) is assigned to experiments with transgenic plants and associated organisms, which, if released outside the greenhouse, could be viable in the surround environment but would have a negligible impact or could be readily managed.

1. Where will the research be conducted?

(Separately list the locations where bench experiments, animal research and plant research will be conducted).

	BUILDING	ROOM NUMBER(S)	FACILITY TYPE	BIOSAFETY LEVEL
Add Row Delete Row			☐ Lab ☐ Animal Facility ☐ Greenhouse	☐ BSL-1 ☐ BSL-2 ☐ ABSL-1 ☐ ABSL-2 ☐ BL1-P ☐ BL2-P

Question 2

Select whether a hand washing sink, eyewash station, or biosafety cabinet is readily available by clicking either the "Yes" or "No" boxes provided. If a biosafety cabinet is available, provide details on the biosafety cabinet's location, model, serial number, and last certification date as requested in the expanding field. To add an additional biosafety cabinet, click the "Add Row" button. Please note that the serial number for the biosafety cabinet may also be a UT Tag number.

c. A biosafety cabinet (BSC)? ☒ YES ☐ NO

	MODEL #	SERIAL #	BUILDING	ROOM #	DATE LAST CERTIFIED
Add Row Delete Row					

Question 3

Include building & room and storage unit descriptions as well as discussion of the measure that will be taken to ensure infectious agents are secured and accounted for. To add an additional row, click the "Add Row" button. Select the appropriate unit type(s) by clicking in the boxes provided. Multiple types of equipment may be checked per specific location. In the last column, please indicate as to whether the equipment and/or room the equipment is located in can be locked by clicking either the "Yes" or "No" boxes provided. If you have selected "No", please describe how the infectious agents are secured and accounted for.

3. Discuss the storage requirements:

(Include building & room and storage unit descriptions as well as discussion of the measure that will be taken to ensure infectious agents are secured and accounted for. Multiple units may be indicated for a single physical location.)

	UNIT TYPE	BUILDING	ROOM #	LOCKABLE?
Add Row Delete Row	☐ -80°C Freezer ☐ Refrigerator / -20°C Freezer ☐ Liquid Nitrogen ☐ Community / Shared Storage ☐ Incubator			☐ YES ☒ NO

If storage units or rooms are not lockable, briefly describe how infectious agents are secured and accounted for:

Question 4

All Personal Protective Equipment (PPE) must be clean and in good repair. This usually necessitates routine cleaning / maintenance for reusable PPE. Methods of cleaning/disinfection and frequency will depend on intended use and risk assessment of biological materials. For more information refer to OSHA PPE standards. Indicate what PPE will be used by checking the appropriate boxes provided.

Question 5

For reusable PPE such as lab coats and eye wear, describe how they will be cleaned or laundered in the expandable text box provided. It is important to note that laundering/cleaning in offsite public or personal home equipment is typically not allowed for PPE worn while handling biohazards or other laboratory hazards (e.g. chemicals, radioactive materials).

Question 6

Indicate which of the following biohazardous sharps will be used by checking the appropriate boxes provided. Also indicate "Yes" or "No" to the questions that appear if you intend to use good sharps practices or require re-capping. If you do require re-capping, visit the Biosafety Office website for a Needle Recapping Exclusion form at: http://biosafety.utk.edu/pdfs/sharps_recap.doc. If biohazardous sharps will not be generated by your research procedures, select the "N/A" box.

6. Indicate which of the following biohazardous sharps will be used: (Check All That Apply) ☐ N/A

☒ glass pipettes ☒ needles ☒ razor blades ☒ scalpel ☒ slides ☒ other

a. Will sharps be managed using good sharps practices including disposal into a biosafety sharps container? ☒ **YES** ☐ **NO**

b. If using needles, do you require re-capping? ☒ **YES** ☐ **NO**

If YES, a Needle Recapping Exclusion form must be on file with the Biosafety Office.
The form is available at: http://biosafety.utk.edu/pdfs/sharps_recap.doc

Question 7

The information provided here should identify the disinfectant product to be used, and how it will be used. Select the appropriate disinfectant(s) listed by active ingredient, and indicate dilution, contact time, and shelf life of each disinfectant. If you select "Other" under Bleach Dilution, specify the dilution to be used in the expandable text box provided.

7. Indicate the surface disinfectant(s) used and provide the specifics for each disinfectant indicated.

	DISINFECTANT	DILUTION	CONTACT TIME	SHELF LIFE
☐	**Bleach**	☐ 1:10 ☒ **Other**		
Please specify bleach dilution:				
☐	**Ethyl / Isopropyl Alcohol**			
☐	**Quaternary Ammonium Compounds**			
☐	**Phenols**			
☐	**Iodine / Iodophoric Solutions**			
☐	**Formaldehyde / Glutaraldehyde**			
☐	**Other:** _____			

Question 8

The *NIH Guidelines* have specific provisions for the IBC to review and approve associated emergency or incident response procedures, including spill response. Contact the Biosafety Office to assist you with developing your spill response plan and to provide guidance for summarizing the information for your registration document.

Question 9

Select all appropriate types of wastes resulting from your research and indicate whether an on-site autoclave, disinfectant, or contractor is used as the method of disposal by clicking in the boxes provided. For on-site autoclaves, indicate the most current validation date, location, and exposure time used. This information can be found posted near any autoclave approved for treatment of bagged biohazardous waste or contacting the Biosafety Office. For disinfectants, indicate the type or dilution and contact time. For medical waste contractors, indicate the name of the contractor company (note the current approved medical waste contractor for all UT-Knoxville area campuses is Stericycle®, Inc.). For "Other" describe how waste will be disposed.

Question 10

Indicate how biohazard(s) will be transported by selecting the appropriate boxes in a-c. Any deviation from the defined methods of transportation should be explained in the expandable text boxes provided. For part d, indicate all regulatory permits that may be applicable to the import, export, or interstate movement of the biological materials described in this registration. Select "N/A" if this section is not applicable to your registration.

10. Indicate how biohazard(s) will be transported (check all that apply):

a. Interlab:	☒	Material will be transported in a sealed leak-proof primary container which will then be placed in a sealed, leak-proof secondary container.

Any deviation should be described below:

b. Interfacility:	☒	Material will be transported in a sealed leak-proof primary container which will then be placed in a sealed, leak-proof secondary container. If transporting in a vehicle, material must be secure.

Any deviation should be described below:

c. Commercial:	☐	Commercial transportation will comply with U.S. Department of Transportation (DOT) or other recognized competent authority. (i.e., ICAO/IATA). Contact the Biosafety Office for commercial shipping assistance.
d. Shipping Permits:		**The following shipping activities might require shipping permits per U.S. Federal Regulations. Please contact the Biosafety Office for more information.**

(Please check all that apply).

- ☐ N/A
- ☐ Import of infectious agents affecting humans; infected human tissues; non-human primate tissues; and vectors of human diseases.
- ☐ Import/Interstate movement of animal pathogens, tissues, or products.
- ☐ Import/Interstate movement of plant pathogens; plant pests; or noxious weeds.
- ☐ Import/Interstate movement of genetically modified or transgenic plants; plant pests; or plant pathogens.
- ☐ Export of infectious agents.
- ☐ Import/Export of wildlife or wildlife samples.

Question 11

This item requires acknowledgement of accident or potential exposure procedures outlined by the university. Actions to take in the event of an exposure include:

1. Flush the exposed area with water. If your eyes, nose or mouth were exposed to blood or other potentially infectious materials, flush these areas for 15 minutes. If your skin was exposed, thoroughly wash these areas with soap and water. Bandage the affected area if needed to control bleeding.
2. Notify your supervisor if he or she is available.
3. Report to the designated medical care provider as soon as possible for follow-up. Take any applicable biological material description documents with you as well.

- For exposure incidents involving human-derived materials (i.e., human cells or blood products), report immediately to UT Medical Center Emergency Room. Identify yourself to ER staff as a UT employee who has had a potential bloodborne pathogens exposure. Contact the Occupational Health Nurse at (865) 974-5728 within 3 days to coordinate further post-exposure evaluation procedures.
- For all other biological material exposures, report as soon as possible to the UT Medical Center or other appropriate medical facility for medical evaluation.
- For any accidents/exposures involving biohazardous materials, notify the Biosafety Office as soon as possible. Both medical evaluation and safety practices follow-up must be completed and documented for such incidents per the provisions of CDC and OSHA standards.

If you answer "NO" or this not applicable to your registration, describe your emergency response procedures in the expandable text box that appears.

Section VI. Personnel

Question 1

List the names and professional credentials of all personnel that will participate in the recombinant DNA research described in this protocol. Professional credentials include job title, degree, date of Standard Microbiological Practices (SMP) training (required for all listed personnel; may be provided by the PI or designate if only BSL-1 work is being performed), the most recent Biosafety Level 2 (BSL-2) training date (if the work will require BSL-2 practices), and the most recent Bloodborne Pathogens (BBP) training date (if work involves personnel handling human derived materials). To add additional rows, click on the "Add Row" button to the left of the table. Note that BSL-2 and BBP training includes Standard Microbiological Practices, so the most recent BSL-2/BBP training date may also be indicated in the SMP field.

Question 2

Describe any health surveillance measures that are required for personnel working on this registration in the expandable text box provided. This may include vaccine offers/requirements, serum surveillance, special precautions for immunocompromised lab personnel, or any other parameters that will be used to ensure/monitor personnel health as determined by a risk assessment of the agents used and procedures performed.

Section VII. Acknowledgement and Approval

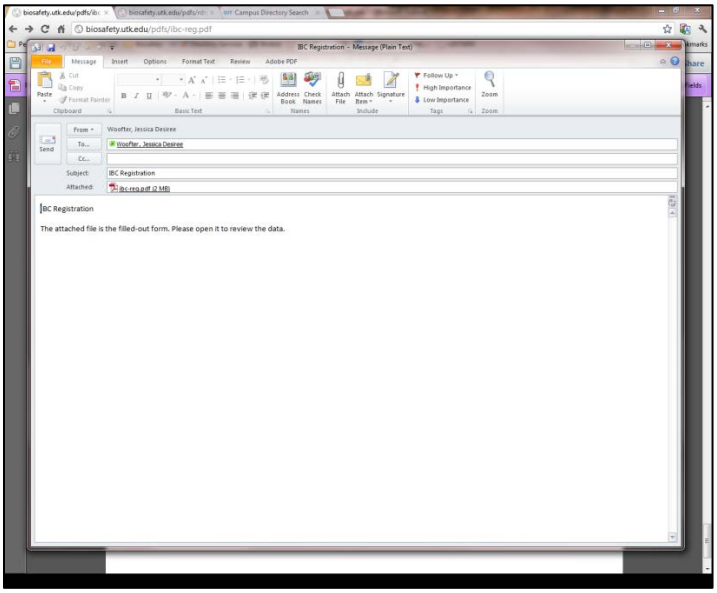
The Principal Investigator and the Department Head must review the applicable statements in this section and provide signatures to acknowledge their responsibilities. These signatures must be provided before final acknowledgment/approval by the Biosafety Officer and IBC Chair upon IBC approval. A copy of the approved registration document with all signatures will be provided to the submitting PI as described previously in this guide.

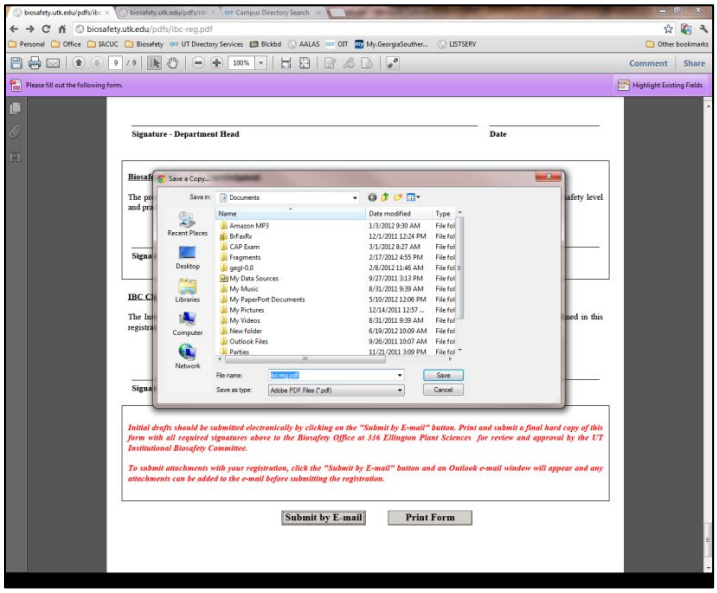
Submitting the Registration

Initial drafts should be submitted electronically by clicking on the "Submit by E-mail" button. This function will also prompt you to save the draft to your computer. I**t is highly recommended that you save the form to your computer for future revisions and processing**. **Failure to save the form may result in lost data, and the form may need to be re-completed in its entirety.**

Once all administrative revisions are completed electronically, print and submit a final hard copy of this form by selecting the "Print Form" button (note that you will be prompted to save after selecting "Print Form" as described above). Submit the final hard copy along with all required signatures to the Biosafety Office at 336 Ellington Plant Sciences for review and approval by the UT Institutional Biosafety Committee.

To submit attachments with your registration, click the "Submit by E-mail" button and an Outlook e-mail window will appear. Any attachments can be added to the e-mail before submitting the registration.





Sample Form

FOR IBC USE ONLY		
IBC Registration #: 001	**Initial Receipt Date:** 5/24/12	**Approval Date:** 6/20/12

Office of Biosafety
2431 Joe Johnson Drive
336 Ellington Plant Sciences Bldg.
Knoxville, TN 37996-4564
Ph: (865) 974-1938 / 974-9836

University of Tennessee Institutional Biosafety Committee Registration

This form is to be used for the registration of research projects involving the use of Recombinant DNA (rDNA), Infectious Agents, Human-Derived Materials, Acute Biological Toxins (LD^{50} < 100 ng/kg) & Select Agent Toxins, Venomous Animals, and/or Poisonous Plants.

All **ORANGE** sections are required and must be completed in their entirety.
Items with text in **BLUE** have hyperlinks that will take you to an external website for additional information.
Items with text in **RED** have additional information that can be viewed by scrolling over the text.
If you require assistance with the form, please call the Biosafety Office at (865) 974-5547.

The completed form must be submitted to the Biosafety Office for evaluation and consultation regarding biological containment. The final registration with all signatures will be submitted to the Institutional Biosafety Committee (IBC) for review and approval. Approvals will be granted for 3 years with annual updates provided through inspections and consultations with the Biosafety Office.

The IBC may request additional registration forms if the proposed project aims are not related.

I. GENERAL INFORMATION

PRINCIPAL INVESTIGATOR

Name: John Jones

Title: Research Assistant Professor Department: Environmental Studies

Mailing Address: 123 Science & Arts

Phone #: 974-0005 E-mail: jonesy@utk.edu

Is there a Co-Investigator or Additional Contact? (Administrative Contact or Person Responsible for Lab)

☐ **NO** ☒ **YES**

	NAME	DEPARTMENT	PHONE #	E-MAIL
Add Row / Delete Row	Jane Doe	Env. Studies	974-0001	jdoe@utk.edu

PROJECT TITLE

Bioluminescent Bioreporter for the Specific Detection of Mercury (II)

Are there any applicable grant titles?

☐ **NO** ☒ **YES**

	GRANT TITLE	FUNDING AGENCY
Add Row / Delete Row	Development of a Bioluminescent Bioreporter for the Specific Detection of Mercury (II)	National Center for Mercury Exposure Prevention

II. NON-TECHNICAL SUMMARY

Provide a non-technical summary of the proposed research for this protocol using language that can be readily understood by non-scientists and reviewers outside of the field of study. Address the purpose/aims of the project and the methods used to conduct this research. Define all acronyms and avoid technical terms.

The objective is to make bioluminescent sensor responsive to mercury (Hg2+). This is accomplished through standard genetic engineering procedures. A genetic element that responds to mercury is fused with genes that mediate bioluminescence. The engineered sensor is inserted into the chromosome of a host microorganism, in this case an innocuous strain of E. coli (EC100). This organism will be used in environmental remediation studies for the rapid detection of free mercury at potentially contaminated sites.

Note: This field is not to exceed 4,000 text characters (~1/2 page).

III. TYPE OF RESEARCH

Please select the appropriate categories for your research registration and complete the sub-sections that follow.

Areas with RED text have additional information that can be viewed by scrolling over the text.
Areas with BLUE text have hyperlinks that will take you to an external website for additional information.

If you need further assistance determining which categories apply to your research, please contact the Biosafety Office.

☒ **RECOMBINANT DNA (rDNA)**

1. Does this project involve any of the following? (Check All That Apply)

A. ☐ Deliberate transfer of drug-resistance traits to microorganisms of clinical, environmental or agricultural significance that could compromise the use of drugs to control disease.

B. ☐ Clinical trial of human gene therapies.

C. ☐ Work with genetic materials from pathogenic microorganisms
(i.e., any agent defined as Risk Group 2 or higher in Appendix B of the *NIH Guidelines*).

D. ☐ Work with a Select Agent or high-consequence pathogen. (i.e., Risk Group 3 or 4 in Appendix B of the *NIH Guidelines*).
You must contact the UTK/UTIA/GSM Biosafety Office immediately for this category!

E. ☐ Work with live animals (i.e., genetic modification of animals or challenge of animals with recombinant DNA or viable recombinant DNA-modified organisms).

F. ☐ Work with live plants (i.e., genetic modification of plants or challenge of plants with recombinant DNA-modified organisms to include microbes and arthropods).

G. ☐ Work involving cloning of genes that encode for toxins

H. ☐ Work that will involve more than 10 liters of culture in one vessel

I. ☒ Not Applicable - Not performing any of the above.

2. Which NIH review classification and category applies to this work?

(Check the appropriate registration category for your experiment AND indicate the subcategory that applies to your work).
Mouse-over or click each NIH category to see the description, to view the full NIH Guidelines go to: http://oba.od.nih.gov/oba/rac/Guidelines/NIH_Guidelines.pdf

REVIEW CATEGORY	SUBCATEGORY
a. ☐	**III-A:** Requires IBC, RAC & NIH approval before initiation

REVIEW CATEGORY		SUBCATEGORY
b.	☐	**III-B:** Requires NIH / OBA and IBC approval before initiation
c.	☐	**III-C:** Requires IBC and IRB approvals and RAC review before participant enrollment. (This category is for human gene transfer protocols only).
d.	☐	**III-D:** Requires IBC approval before initiation
e.	☒	**III-E:** Requires IBC notice simultaneous with initiation Subcategory: E
f.	☐	**III-F:** IBC registration is not required but recommended to assure that work is properly classified

3. List all hosts, vectors and inserts used as part of this protocol in the table below.
*Give specific strains, promoter names, gene families, or class at a minimum. Catchalls like "..and other genes" are **NOT** acceptable.*

A detailed description of recombinant construct development must be given in Section IV - Technical Summary.

HOSTS	E. coli EC100 competent cells (Epicenter, Madison, WI) E. coli DH5α
VECTORS	pDG106 - contains the mer operon Plasmid pFSP3 - EZ::TN containing the promoterless lux cassette
PROMOTERS	PmerT
INSERT GENES	Final chromosomal insert merR PmerT luxCDABE Phenotype is production of bioluminescense in the presence of Hg2+

☐ **INFECTIOUS AGENTS**

☐ **HUMAN-DERIVED MATERIALS**

☐ **ACUTE BIOLOGICAL TOXINS (LD^{50} < 100 ng/kg) & SELECT AGENT TOXINS**

☐ **VENOMOUS ANIMALS**

☐ **POISONOUS PLANTS**

IV. TECHNICAL SUMMARY

Provide a technical description of the proposed research for this registration. Address the purpose and aims of the project including a brief but complete description and the methods used to conduct this research. Define all acronyms and include sufficient detail for the committee to make a risk assessment of the project.

Descriptions of recombinant DNA work must include information that supports the review category of this research as classified under the NIH Guidelines and must reflect the materials identified for Section III - Recombinant DNA- Question 3 (Hosts, Vectors, Promoters, Insert Genes Table).

A 505 bp merR fragment was PCR amplified from the mer operon and cloned into the TA Cloning Vector (pCR2.1; Invitrogen, San Diego, CA). Primers (Table 1) for the amplification were synthesized based on the merRo/p sequence listed in GenBank (Accession #AF071413; nucleotides 19133-19638). The source of the mer DNA was pDG106 (Gambill and Summers, 1985).

The merR was excised from pCR2.1-merR with EcoRV and BamHI, ligated into plasmid pFSP3 (transposon vector containing luxCDABE), and transformed into chemically competent E. coli DH5α using standard molecular techniques. Transformants were subjected to miniprep plasmid isolation and further screened by restriction digestion with BamHI and KpnI. A positive clone designated #7 contained the merR gene in the proper orientation to induce bioluminescence in the presence of Hg2+ ions.

Following large-scale preparation, the transposon vector was digested with PshAI overnight at 25ºC. The 8.5 kb fragment containing the mer-lux reporter transposon was gel purified with Gene Clean. The transposome was formed by incubating the 8.5 kb fragment (mer lux EZ::TN) with transposase according to manufacturer's directions. The resultant transposome was then electroporated into E. coli EC100 competent cells (Epicenter, Madison, WI).

Electroporants were plated on LB agar plates with Km (50 mg/L). Three colonies were recovered which produced bioluminescence in the presence of Hg2+ ions. These strains were designated E. coli ARL1, E. coli ARL2, and E. coli ARL3 and will be used for screening of mercury-containing environmental samples.

Note: Add additional resources pertaining to the technical summary as an e-mail attachment along with this registration. This field is not to exceed 10,000 text characters (~2 pages).

V. FACILITIES & PROCEDURES ASSESSMENT

1. Where will the research be conducted?

(Separately list the locations where bench experiments, animal research and plant research will be conducted).

	BUILDING	ROOM NUMBER(S)	FACILITY TYPE	BIOSAFETY LEVEL
Add Row / Delete Row	Calvin Science & Engineering (CSE)	2212	☒ Lab ☐ Animal Facility ☐ Greenhouse	☒ BSL-1 ☐ BSL-2 ☐ ABSL-1 ☐ ABSL-2 ☐ BL1-P ☐ BL2-P

2. Are the following readily available:

a. A hand washing sink? ☒ **YES** ☐ **NO**

b. An eyewash station? ☒ **YES** ☐ **NO**

c. A biosafety cabinet (BSC)? ☒ **YES** ☐ **NO**

	MODEL #	SERIAL #	BUILDING	ROOM #	DATE LAST CERTIFIED
Add Row / Delete Row	TX123	123456	CSE	2212	3/7/12

3. Discuss the storage requirements:

(Include building & room and storage unit descriptions as well as discussion of the measure that will be taken to ensure infectious agents are secured and accounted for. Multiple units may be indicated for a single physical location.)

	UNIT TYPE	BUILDING	ROOM #	LOCKABLE?
Add Row / Delete Row	☒ -80°C Freezer ☒ Refrigerator / -20°C Freezer ☐ Liquid Nitrogen ☐ Community / Shared Storage ☒ Incubator	CSE	2212	☒ YES ☐ NO

4. Indicate the Personal Protective Equipment (PPE) that will be used.

Note: All PPE must be clean and in good repair. All reusable PPE will require routine cleaning / maintenance. Methods of cleaning / disinfection and frequency will depend on intended use and risk assessment of biological materials. For more information refer to OSHA PPE standards.

☐ **Gowns** ☒ **Lab Coats** ☐ **Shoe Covers** ☐ **Booties**

☒ **Gloves** ☒ **Safety Glasses** ☐ **Respirators** ☐ **Goggles**

5. For reusable Personal Protective Equipment (PPE) such as lab coats & eye wear, please describe how they will be cleaned or laundered:

Lab coats will be laundered in the designated departmental laundering facility, and eye wear will be disinfected with 1:10 bleach solution and washed with soap and water.

6. Indicate which of the following biohazardous sharps will be used: (Check All That Apply) ☐ N/A

☒ glass pipettes ☐ needles ☐ razor blades ☐ scalpel ☒ slides ☒ other

a. Will sharps be managed using good sharps practices including disposal into a biosafety sharps container? ☒ **YES** ☐ **NO**

b. If using needles, do you require re-capping? ☐ **YES** ☒ **NO**

7. Indicate the surface disinfectant(s) used and provide the specifics for each disinfectant indicated.

	DISINFECTANT	DILUTION	CONTACT TIME	SHELF LIFE
☒	**Bleach**	☒ **1:10** ☐ **Other**	15 minutes	1 week
☐	**Ethyl / Isopropyl Alcohol**			
☐	**Quaternary Ammonium Compounds**			
☐	**Phenols**			
☐	**Iodine / Iodophoric Solutions**			
☐	**Formaldehyde / Glutaraldehyde**			
☐	**Other:** _____			

8. Summarize your written biological spill response procedure for the materials covered by this registration which has been reviewed by the UT Biosafety Officer. (Contact the UTK/UTIA/GSM Biosafety Officer for assistance at (865) 974-1938). Sample emergency responses available at: **http://biosafety.utk.edu/emergency/**

For spill response, wear gloves, a lab coat and eye protection. Splash goggles are required for bleach solution preparation. Be careful not to "track" spills with your feet. If the spill has affected a large area of the floor, wear shoe covers or disinfect the soles of your shoes if you have to walk through contamination for spill cleanup.
If the spill does not involve any broken glass or other sharps, place paper towels over the spill. Using freshly prepared 1:10 bleach solution, spray disinfectant on the paper towels beginning outside the spill and work inward to the center of the spill area. Remove the spill and saturated paper towels. Apply disinfectant to the affected surfaces and allow a 15 minute contact time before wiping up any residues. Bleach residues can be further reduced by wiping down the area with a mild detergent or ethanol. Dispose of wastes in the biohazardous waste container.
If the spill involves sharps or broken glass, remove as much of this as possible using forceps, tongs or cardboard wedges. Place the broken glass/sharps in an autoclavable bucket or sharps container. Apply disinfectant and absorbent powder to the affected area and then sweep up the spill with a disposable broom and dust pan. Place the wastes in the biohazardous waste container. Apply fresh disinfectant to the affected surfaces and allow a 15 minute contact time before wiping up any residues. Bleach residues can be further reduced by wiping down the area with a mild detergent or ethanol. (Note: if the spill involved broken glass, use a mechanical device to "wipe up" the spill- avoids wiping surfaces by hand.)

9. Type of waste and disposal method: (Check all types of wastes generated by your research procedures).

	TYPE OF WASTE	METHOD OF DISPOSAL
☒	**Liquids**	
	☒ On-Site Autoclave:	Location: CSE 2110
	☐ Disinfectant:	
☒	**Solid Wastes, Non-Sharps**	
	☒ On-Site Autoclave:	Last Validation Date: 4/26/12 Exposure Time: 45 minutes
		Location: CSE 2110
	☐ Medical Waste Contractor:	
	☐ Other:	
☒	**Biohazardous Sharps**	
	☒ Medical Waste Contractor:	Contractor: Stericycle, Inc.
	☐ Other:	
☐	**Animal Carcasses / Pathological Waste**	
☐	**Toxin**	

10. Indicate how biohazard(s) will be transported (check all that apply):

a. Interlab:	☒	Material will be transported in a sealed leak-proof primary container which will then be placed in a sealed, leak-proof secondary container.

Any deviation should be described below:

As indicated

b. Interfacility:	☐	Material will be transported in a sealed leak-proof primary container which will then be placed in a sealed, leak-proof secondary container. If transporting in a vehicle, material must be secure.
c. Commercial:	☒	Commercial transportation will comply with U.S. Department of Transportation (DOT) or other recognized competent authority. (i.e., ICAO/IATA). Contact the Biosafety Office for commercial shipping assistance.
d. Shipping Permits:		**The following shipping activities might require shipping permits per U.S. Federal Regulations. Please contact the Biosafety Office for more information.**

(Please check all that apply).

☒ N/A

11. Accidents, Exposures, & Emergency Response

☒ **YES** ☐ **NO**

In the event of an accident/potential exposure, do you agree to follow the procedures listed below?

Actions to take in the event of an exposure.....

1. Flush the exposed area with water.
If your eyes, nose or mouth were exposed to blood or other potentially infectious materials, flush these areas for 15 minutes. If your skin was exposed, thoroughly wash these areas with soap and water. Bandage the affected area if needed to control bleeding.

2. Notify your supervisor if he or she is available.

3. Report to the designated medical care provider as soon as possible for follow-up. Take any applicable biological material description documents with you as well.

- For exposure incidents involving human-derived materials (i.e., human cells or blood products), report **immediately** to UT Medical Center Emergency Room. Identify yourself to ER staff as a UT employee who has had a bloodborne pathogens exposure. Contact the Occupational Health Nurse at (865) 974-5728 within 3 days to coordinate further post-exposure evaluation procedures.

- For all other biological material exposures, report as soon as possible to the UT Medical Center or other appropriate medical facility for medical evaluation.

- For any accidents/exposures involving biohazardous materials, notify the Biosafety Office as soon as possible. Both medical evaluation and safety practices follow-up must be completed and documented for such incidents per the provisions of CDC and OSHA standards.

VI. PERSONNEL

1. List all personnel who will be working under this registration, their expertise and training:

	NAME	TITLE	DEGREE	TRAINING DATES
Add Row / Delete Row	John Jones	Assistant professor	PhD	SMP: 2/5/05 BSL-2 : BBP:
Add Row / Delete Row	Jane Doe	Research Associate	MS	SMP: 1/31/06 BSL-2 : BBP:
Add Row / Delete Row	William Wright	Graduate Student	BS	SMP: 1/31/06 BSL-2 : BBP:

2. Describe any health surveillance measures that are required for personnel working on this registration.
(i.e., vaccination offers, health conditions that could increase exposure or disease risk, etc.**)**

All E. coli strains are Risk Group 1 and will not cause disease in healthy personnel. Those with immune deficiencies or underlying health concerns are asked to self-identify so additional precautions can be taken if necessary.

VII. ACKNOWLEDGEMENT AND APPROVAL

Principal Investigator's Acknowledgment of Regulatory Requirements

As the Principal Investigator for this registration, I understand and acknowledge my responsibilities to:

- Abide by the provisions of the *NIH Guidelines*, the *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, and/or the (T)OSHA/ Bloodborne Pathogen (BBP) Standard as applicable;
- Comply with UT Biosafety Policies and Procedures as well as local, state, and federal regulations;
- Be adequately trained in good microbiological techniques (including standard microbiological practices) and ensure that all personnel working on this protocol are also trained in these techniques;
- Make available to all laboratory staff the protocols that describe the potential biohazards and the precautions to be taken;
- Inform the laboratory staff of the reasons and provisions for any precautionary medical practices advised or requested (e.g., vaccinations or serum collection);
- Supervise the safety performance of the laboratory staff to ensure that the required safety practices and techniques are employed;
- Ensure the integrity of the physical containment (e.g., biological safety cabinets) and the biological containment (e.g., purity and genotypic and phenotypic characteristics);
- Investigate and report any significant problems pertaining to the operation and implementation of containment practices and procedures in writing to the Biological Safety Officer (where applicable), Greenhouse/Animal Facility Director (where applicable), IBC, NIH/OBA, and other appropriate authorities (if applicable);
- Adhere to IBC-approved emergency plans for handling accidental spills and personnel contamination;
- Make available lab protocols including information regarding potential biohazards and the precautions to be taken to the laboratory staff;
- Ensure that research personnel are trained in: standard microbiological practices and techniques required to ensure safety for this project, emergency response procedures, exposure incident response procedures, waste management procedures, and the requirements of the OSHA Bloodborne Pathogens Standard (as applicable).
- Supervise staff and correct work errors and conditions that could result in breaches of biosafety and/or biological containment practices prescribed for this work.

Signature - Principal Investigator **Date**

Department Head's Acknowledgment

I have reviewed this document and will ensure the Faculty/Principal Investigator will carry out their work in accordance with applicable health & safety regulations and guidelines. I agree to contact the Biosafety Office if the Principal Investigator cannot fulfill the responsibilities outlined in this registration.

Signature - Department Head **Date**

Biosafety Officer's Acknowledgment

The procedures and facilities have been evaluated in conjunction with the Principal Investigator and the proposed biological safety level and practices are appropriate for the proposed use of the biological materials outlined in this document.

Signature - Biosafety Officer **Date**

IBC Chair's Acknowledgment

The Institutional Biosafety Committee has approved the biological materials, safety practices, and containment facilities outlined in this registration.

Signature - IBC Chair **Date**