



Institutional Biosafety Committee
Office of Research Affairs, Research Administration Bldg.
University of Massachusetts Amherst
70 Butterfield Terrace, Amherst, MA 01003-9242
Telephone: (413) 545-5283; Fax: (413) 577-1728

UNIVERSITY OF MASSACHUSETTS AMHERST

Registration Document for Recombinant DNA Research

The University of Massachusetts Amherst is required by law to ensure that all research activities involving recombinant DNA (rDNA) technology are reviewed in accordance with the NIH Guidelines. This includes unfunded projects, projects that are funded by an agency other than NIH.

An rDNA registration must be submitted to the University of Massachusetts Amherst's Institutional Biosafety Committee (IBC) for review to ensure that all provisions of the NIH Guidelines will be followed. Please complete and submit the attached rDNA Registration Form to Melinda Lalecheur in the Office of Research Affairs (413-545-5275) for review to determine the NIH review level. rDNA research at UMass Amherst generally falls into one of three NIH review levels – IIID (IBC review and approval required before the work starts), IIIE (notify the IBC before the work starts, IBC review required) and IIIF (exempt from IBC oversight). If your research is review level IIIF you will be asked to confirm its exempt status each time you submit a new grant proposal to a funding agency, including non-competing renewals, and must report changes in your procedures that could change the review level. For specific questions about managing rDNA research please contact the campus Biological Safety Services Manager at jladuc@ehs.umass.edu at 413-545-7293.

An rDNA registration may be approved for up to 5 years. A new registration renewal must be submitted to extend the approval period.

General Instructions:

Please complete:

- Page 2 Principal Investigator and Contact Information
- Section 1. General Project Description
- Section 2. Recombinant DNA and rDNA Exempt Details
- Section 3. Dual Use Questionnaire
- Section 4. Principal Investigator Certification

Principal Investigator and Contact Information

Responsible PI:

Name: _____

Department: _____

Building: _____

Lab Room Number(s): _____

Office Phone: _____ Lab Phone _____

E-Mail: _____ Home Phone: _____

Provide the name and contact information for an alternate contact who can either answer questions for the IBC or an EH&S Inspector and/or who will always be able to contact you.

Alternate Contact:

(A Senior Research Assistant, Lab Manager or Co-PI, who is informed of the lab's research protocols and safety and emergency response procedures)

Lab Contact _____

Department: _____

Building: _____

Room Number: _____

Office Phone: _____ Lab Phone _____

E-Mail: _____ Home Phone: _____

Section 1. General Project Description

(to be completed for all applications)

Project Title:

Project Period: to

Funding Agency:

General Description

Briefly describe the research proposed that involves the use of rDNA technology. Please write your description so that non-scientists on the IBC can understand how you propose to use rDNA. Please avoid or define acronyms and abbreviations. Be sure to describe the origin or source of DNA and the host – vector system you will use. Include a flow chart for complex projects.

Section 2. Recombinant DNA (rDNA)

(To be completed for rDNA applications only)

Please identify any of the following that you intend to use in your research:

- Does the work involve transfer of a drug resistance trait to an organism that does not acquire it normally (Check "no" for standard drug resistance, e.g., ampicillin into *E. coli*)? Yes No
- Does the rDNA contain genes coding for molecules toxic to vertebrates (LD50 <100 nanograms / kg body wt)? Yes No
- Will the rDNA be used in human subjects (human gene transfer experiments)? Yes No
- Do the rDNA experiments involve human genes being cloned into non-pathogenic prokaryotic or lower eukaryotic host-vector systems? Yes No
- Will there be any large-scale experiments (i.e. requiring more than 10 liters of culture)? Yes No
- Will genes from one microbe be used to express in another? Yes No
- Are any human or animal pathogens used either as the host organism or as a vector? Yes No
- Is any DNA from Risk Group 2, 3, or 4 agents or restricted organisms cloned into non-pathogenic prokaryotic or lower eukaryotic host-vector systems? Yes No
- Do rDNA or RNA experiments involve the use of infectious animal or plant viruses in tissue culture systems or in animals? Yes No
- Do rDNA or RNA experiments involve the use of defective animal or plant viruses in the presence of helper virus in tissue culture systems or in animals? Yes No
- Do any rDNA molecules contain one half or more of any eukaryotic viral genome? Yes No
- Do rDNA experiments involve whole animals? Yes No
- Do rDNA experiments involve whole plants? Yes No
- Will you release rDNA into the environment? Yes No
- Will you be breeding transgenic animals to create novel genetically engineered animals? Yes No

If you answered NO to all of the questions, your rDNA research may be exempt from the NIH Guidelines i.e. not require full review by the Institutional Biosafety Committee. If you believe your project is exempt please indicate which of the exemptions to the NIH Guidelines listed on the next page applies to your research.

Section 2. (Continued) rDNA Exempt Categories

To determine whether your project and host-vector system are exempt under NIH Guidelines refer to the complete text of Section III Experiments Covered by the NIH Guidelines:

http://www4.od.nih.gov/oba/rac/guidelines_02/NIH_Guidelines_Apr_02.htm#_Toc7261559

Exempt recombinant DNA molecules [[Section III-F](#) of the NIH Guidelines]:

- III-F-1 are NOT in organisms and viruses, or
- III-F-2 consist entirely of DNA segments from a single non-chromosomal or viral DNA source, or
- III-F-3 consist entirely of DNA from a prokaryotic host including its indigenous plasmids or viruses when propagated only in that host or when transferred to another host by well established physiological means, or
- III-F-4 consist entirely of DNA from a eukaryotic host including its chloroplasts, mitochondria, or plasmids when propagated only in that host or a closely related strain of the same species, or
- III-F-5 consist entirely segments from different species that exchange DNA by known physiological processes, though one or more may be a synthetic equivalent (see [Appendix A](#) of the NIH Guidelines), or
- III-F-6 do NOT present a significant risk to health or the environment as determined by the NIH Director (see [Appendix C](#) of the NIH Guidelines) and as summarized below (*on next page*):
 - C-I **Recombinant DNA in Tissue Culture** [[Appendix C-I](#)]. Recombinant DNA molecules containing less than one-half of any eukaryotic viral genome (all viruses from a single family being considered identical) are exempt unless there is an applicable exception. That is, the total of all eukaryotic viral genomes within a Family shall not exceed one-half the genome. Is there is an applicable exception that voids this exemption (note [Appendix C-I-A](#))?
 No Yes. If yes, what is the number of the exception? _____
 - C-II **Escherichia coli K-12 Host-Vector Systems** [[Appendix C-II](#)]. Experiments that use E. coli K-12 host-vector systems are exempt unless there is an applicable exception.
Is there an exception that voids this exemption (note [Appendix C-II-A](#))?
 No Yes. If yes, what is the number of the exception? _____
 - C-III **Saccharomyces Host-Vector Systems** [[Appendix C-III](#)]. Experiments that use *Saccharomyces cerevisiae* and *S. uvarum* host-vector systems are exempt unless there is an applicable exception.
Is there an exception that voids this exemption (note [Appendix C-III-A](#))?
 No Yes. If yes, what is the number of the exception? _____

- C-IV ***Bacillus subtilis* or *B. licheniformis* Host-Vector Systems** [[Appendix C-IV](#)]. Any asporogenic or asporogenic strain which does not revert to a spore-former with a frequency greater than 10^{-7} may be used for cloning DNA unless there is an applicable exception.
Is there an exception that voids this exemption (note [Appendix C-IV-A](#))?
 No Yes. If yes, what is the number of the exception? _____
- C-IV **Extrachromosomal Elements of Gram Positive Organisms** [[Appendix C-V](#)]. rDNA molecules derived entirely from extrachromosomal elements of the organisms listed in Appendix C-V (including shuttle vectors constructed from vectors described in Appendix C), propagated and maintained in organisms listed in Appendix C-V are exempt unless there is an applicable exception.
Is there an exception that voids this exemption (note [Appendix C-V-A](#))?
 No Yes. If yes, what is the number of the exception? _____
- C-V **Transgenic Rodents** [[Appendix C-VI](#)]. The purchase or transfer of transgenic rodents for experiments that require Biosafety Level 1 [[Appendix G-III-M](#)] containment.

Section 3. Dual Use Technologies

Does your research involve any experiments that would?

1. Demonstrate how to render a vaccine ineffective. Yes No
2. Render a pathogen (\geq risk group 2) resistant to antibiotics or antivirals Yes No
3. Enhance a pathogen's virulence or render a non-pathogen virulent Yes No
4. Increase a replication competent pathogen's transmissibility Yes No
5. Alter a replication competent pathogen's host range Yes No
6. Enable a pathogen to evade diagnostic tests Yes No
7. Enable weaponization of pathogens and toxins Yes No

Section 4. Principal Investigator Certification

By signing below, I certify that I have read the following statements and agree to abide by them and other UMass policies and procedures governing the use of recombinant DNA, infectious agents and other biological materials, as outlined in this application and in the following:

I will:

- a) Ensure that listed personnel have received biological safety training (<http://www.ehs.umass.edu/Biological%20Safety%20Training.htm>) in safe laboratory practices and the procedures for this protocol *before any work begins on this project*, received follow-up trainings as required by University policy, and that all personnel who could have occupational exposure to bloodborne pathogens have received appropriate bloodborne pathogen training.
- b) Follow the health surveillance practices outlined by the biosafety officer for this protocol.
- c) Report accidental exposures or releases to Judy LaDuc, Biological Safety Services Manager (EH&S) at 413-545-7293. Any releases must be reported to the NIH, therefore I must notify EH&S so that the appropriate action is taken.
- d) Have myself or my staff report to UHS (or an Emergency Room) to be seen by medical staff and inform EH&S at 413-545-2682 of any research-related accident or illness as soon as possible after its occurrence.
- e) Submit a request for approval to the IBC for any significant modifications to the study, facilities or procedures. This request should be submitted to Hilary Woodcock hilaryw@ora.umass.edu and it will be reviewed by the Biological Safety Officer as well as other members of the IBC as deemed necessary.
- f) Comply with safety practices as described in the most recent version of the Biosafety in Microbiological and Biomedical Laboratories² (<http://www.cdc.gov/od/ohs/biosfty/bmb15/bmb15toc.htm>) for work with pathogens.
- g) Ensure that all proposed live vertebrate animal use has been approved by the UMass Institutional Animal Care and Use (IACUC).
- h) Ensure that all biohazardous waste and/or sharps waste is decontaminated and handled in accordance with EH&S medical/biological waste guidelines (<http://www.ehs.umass.edu/bio.htm>). Recombinant DNA waste must be autoclaved prior to disposal. Animal carcasses are frozen for at least two days then packaged in bio-boxes for incineration. Sharps containers and bio-boxes are removed for incineration by completing the form on this website: <http://www.umass.cems.sr.unh.edu/CEMS/RequestRemoval>
- i) Comply with NIH requirements pertaining to the shipment and transfer of recombinant DNA materials¹.
- j) Become familiar with, and abide by, all provisions of the most current NIH Guidelines¹.
Appendix H - Shipment: http://oba.od.nih.gov/oba/rac/guidelines_02/NIH_Gdlnes_Ink_2002z.pdf

Signature: _____
(Principal Investigator)

Date: _____

1. NIH recombinant DNA Guidelines <http://www4.od.nih.gov/oba/rac/guidelines/guidelines.html>
2. CDC / NIH publication Biosafety in Microbiological and Biomedical Laboratories
<http://www.cdc.gov/od/ohs/biosfty/bmb15/bmb15toc.htm>
3. OSHA Bloodborne Pathogen Standard:
http://www.osha.gov/pls/oshaweb/owadisp.show_document?p_table=STANDARDS&p_id=10051

When completed, please return this form to:

Melinda Lelacheur, Program Administrator,
Office of Research Affairs, Research Administration Building,
University of Massachusetts Amherst, 70 Butterfield Terrace
Amherst, MA 01003-9242
Telephone: (413) 545-5283
Fax: (413) 577-1728

<i>****For Institutional Biosafety Committee (IBC) Use Only****</i>					
<input type="checkbox"/> Approve	<input type="checkbox"/> Approve with Stipulations	<input type="checkbox"/> Disapprove	<input type="checkbox"/> Exempt from IBC Oversight		
NIH Level:	<input type="checkbox"/> III-A	<input type="checkbox"/> III-B	<input type="checkbox"/> III-C	<input type="checkbox"/> III-D	<input type="checkbox"/> III-E <input type="checkbox"/> III-F
Signature: _____	Date: _____				

Recombinant DNA/Infectious Agent Registration

Below is a Summary of the Registration Requirements described in the NIH Guidelines for Research Involving Recombinant DNA Molecules (<http://www4.od.nih.gov/oba/>).

Level	Approval/Review	Requirements
III-A	NIH Director, RAC, IBC	A drug resistant gene transferred into a (new) microorganism.
III-B	NIH/OBA, IBC	The cloning of toxin molecules with LD ₅₀ < 100 ng/kg of body weight.
III-C	RAC, IRB, IBC	Recombinant DNA (or DNA or rDNA derived from rDNA) transferred into humans.
III-D	IBC [†]	Recombinant DNA transferred to or from whole animals, whole plants, transgenic rodents, experiments involving >10 Liters of culture, or agents listed in Risk Groups 1, 2, 3, or 4 (see below) at the appropriate Biological Safety Level (BSL).
III-E	IBC [§]	Recombinant DNA involving no more than 2/3 eukaryotic virus agents, whole plants, arthropods, or transgenic rodents.
III-F		Recombinant DNA not found in organisms or viruses, single monochromal or viral DNA sources, or host DNA transferred to the same host or related species.

[†] Approval required before initiation.

[§] Notify IBC when project is initiated. IBC approval still required.

Risk Groups ^λ	
Risk Group 1 (RG1)	Agents that are not associated with disease in healthy adult humans. (often BSL-1)
Risk Group 2 (RG2)	Agents that are associated with human disease which is rarely serious and for which preventive or therapeutic interventions are often available. (often BSL-2)
Risk Group 3 (RG3)	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). (often BSL-3)
Risk Group 4 (RG4)	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). (often BSL-4)

^λ For a listing of agents, see “Appendix B – Classification of Human Etiologic Agents on the Basis of Hazard” in the NIH Guidelines For Research Involving Recombinant DNA Molecules, April 2002.



1. Under which section of the *NIH Guidelines* does the generation of transgenic rodents fall?

The creation of transgenic rodents falls under one of two portions of the *NIH Guidelines* depending on the containment level required to house the rodents. Experiments involving the creation of transgenic rodents that can be housed under Biosafety Level 1 conditions are covered under Section III-E-3. Experiments involving the generation of transgenic rodents requiring BL2, BL3 and BL4 containment are covered under Section III-D-4.

2. Under which section of the *NIH Guidelines* does the generation of transgenic animals other than rodents fall?

The creation of all transgenic animals (other than rodents that can be housed under BL1 containment conditions) is covered under Section III-D-4 of the *NIH Guidelines*.

3. Would the breeding of two different strains of knock-out mice require IBC approval under the *NIH Guidelines*?

The techniques used initially to create knock-out animals involve the stable introduction of recombinant DNA into the animal's genome, and thus these animals are considered transgenic. As the breeding of two different strains of knock-out mice will potentially generate a novel strain of transgenic animal, the work is covered under the *NIH Guidelines* and as such requires IBC review and approval. Sections in the *NIH Guidelines* that cover work with rodents include III-E-3 for work that requires Biosafety Level (BL) 1 containment and III-D-4 for work that requires BL2, BL3 and BL4 containment.

4. Is IBC registration and approval needed for the maintenance of a transgenic animal colony?

The maintenance of a transgenic rodent colony (i.e. breeding within a particular transgenic strain) at BL1 is an activity that is exempt from the *NIH Guidelines* and, as such, does not require IBC registration and approval. The maintenance of a transgenic rodent colony at BL2 or higher falls under Section III-D-4-b and requires IBC approval. The breeding of all other transgenic animals is subject to the *NIH Guidelines* under Section III-D-4-a or III-D-4-b depending on the containment level required.

5. Is the purchase and transfer of transgenic rodents exempt from the *NIH Guidelines*?

Under Appendix C-VI of the *NIH Guidelines*, the purchase or transfer of transgenic rodents may be maintained at BL1 containment are exempt from the *NIH Guidelines*. The purchase or transfer of transgenic rodents that require BL2 or higher containment is not exempt from the *NIH Guidelines*. These animals are covered under Section III-D-4, and purchase and transfer of such animals requires IBC registration and approval.

It should be noted that the subsequent use of transgenic rodents may not be exempt from the *NIH Guidelines*. Experiments using transgenic rodents at BL1 are exempt from the *NIH Guidelines* if the experiment does not involve the use of recombinant DNA. If the protocol does involve the use of recombinant DNA or is conducted at BL2 or higher then the work falls under III-D-4 of the *NIH Guidelines* and as such requires IBC review and approval prior to initiation.

6. Is the purchase and transfer of transgenic animals other than rodents exempt from the *NIH Guidelines*?

No, only the purchase or transfer of transgenic rodents that may be maintained at BL1 containment is exempt from the *NIH Guidelines*. The purchase or transfer of any other animal for research purposes at any biosafety level (including BL1) is not exempt, nor is the purchase and transfer of transgenic rodents that require BL2 or higher containment.

7. Are gene ablation studies covered by the NIH Guidelines?

The answer to this question depends on the technique employed in the study. If recombinant techniques are used to knock out the gene, then work would be covered under the *NIH Guidelines*.

8. Who has the responsibility to review the generation of transgenic animals if an institution is generating animals for investigators who are not affiliated with that institution?

The generation (creation) of transgenic animals is an activity covered under the *NIH Guidelines*. The IBC at the institution where that activity is occurring has the responsibility to review and approve that activity (if the institution is subject to the requirements of the *NIH Guidelines*). The subsequent use of the animals by investigators not at that institution would need to be reviewed and approved by the IBC at the investigator's institution if that institution conducts or supports recombinant DNA research that receives NIH support and the activity covered under the *NIH Guidelines*.

9. When a core facility generates transgenic mice as a "fee for service" for Principle Investigators (PIs), is it the responsibility of the PI or the core facility to register the generation of the mice with the IBC?

Section IV-B-7-a-(1) of the *NIH Guidelines* articulates one of the responsibilities of the PI as 'initiating no recombinant DNA research which requires IBC approval prior to initiation until that research has been approved by the IBC and has met all other requirements of the *NIH Guidelines*.' It would be acceptable for either the PI of the core facility or the PI purchasing the transgenic animals to fulfill the responsibility to register the generation of the animals. In many cases, the animals being generated will be subsequently used in experiments that are subject to the *NIH Guidelines*, and the registration of the research with the IBC may encompass both the generation and subsequent experimentation with the animals.

10. When existing transgenic animals at an institution are purchased or transferred to an investigator outside the institution, who should review and approve the use of these animals?

An institution's IBC does not need to review and approve the use of transgenic animals at another institution. If the receiving institution is subject to the *NIH Guidelines* (i.e. conducts or supports recombinant DNA research that receives NIH support), then the purchase and transfer of animals (other than rodents that can be housed under BL1 containment), along with any experiments subject to the *NIH Guideline*, would require review and approval by the IBC at that institution.

11. What are the NIH Guidelines requirements for research with large transgenic animals (sheep, pigs, etc.), or research with recombinant DNA microorganisms in such animals?

When conducting recombinant DNA work with large animals, the work is covered under Appendix Q of the *NIH Guidelines*. Appendix Q specifies containment and confinement practices when animals are of a size or have growth requirements that preclude the use of laboratory containment of animals. The *NIH Guidelines* include provisions for tracking and inventorying these animals (Appendix Q-1-B-2 states that a permanent record must be maintained of the experimental use and disposal of each animal). Animal carcasses must be disposed of as to avoid their use as food for human beings or animals unless food use is specifically authorized by an appropriate federal agency (Appendix Q-1-B-1). An acceptable method, for example, would be incineration.

12. Are recombinant DNA modifications to the somatic cells of non-transgenic animals subject to the NIH Guidelines?

Yes, these experiments are subject to the *NIH Guidelines*.

- a) Sections III-D-1-a through III-D-1-d cover experiments using Risk Group 2, 3, 4, or restricted agents in whole animals. See the *NIH Guidelines* for the appropriate containment for such experiments
- b) Section III-D-4-a covers experiments involving viable recombinant DNA-modified microorganisms tested on whole animals. DNA from any source except for greater than two-thirds of eukaryotic viral genome may be transferred to any animal and propagated under conditions of physical containment comparable to BL1 or BL1-N and appropriate to the organism under study.
- c) Section III-D-4-b covers recombinant DNA, or DNA or RNA derived therefrom, involving whole animals, including transgenic animals that are not covered by Sections III-D-1 or III-D-4-a. The appropriate containment for these experiments is determined by the IBC.
- d) Experiments not included in Sections III-A, III-B, III-C, III-D, III-F, fall into Section III-E. Experiments covered by Section III-E may be conducted at BL1 containment.