



I. Purpose

To ensure that animal facility security, animal containment, and animal disposal is being conducted according to the [NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules](#) (NIH Guidelines, 2013).

This policy describes Principal Investigator responsibilities, institutional review and approval procedures, and animal containment and disposal procedures required for all research or teaching activities involving animals in which the animal's genome will be altered by introduction of recombinant DNA into the germ-line (production of transgenic, knock-out and knock-in animals).

Of note, research involving recombinant or synthetic nucleic acid molecules in large animals is not currently permitted at Dartmouth College. This SOP is developed for small animal work only (rodents, ferrets).

I. Regulatory Background

- All institutions that receive NIH funding for any research involving recombinant or synthetic nucleic acids (r/sNA) must comply with the *NIH Guidelines*.
- Experiments involving transgenic vertebrate animals are subject to *NIH Guidelines* Section III-D-4, III-E, and Appendix Q. All transgenic organisms must be rendered biologically inactive by appropriate methods before disposal outside the facility (Appendix G-II-B-2-i and G-II-C-2-n).
- Dartmouth College Institutional Biosafety Committee (IBC) requires that all research involving transgenic animals must receive approval from the IBC prior to commencement according to the procedures outlined in this SOP.
- **NOTE: the purchase or transfer of commercial whole transgenic rodents is exempt from IBC review under the NIH Guidelines under Section III-D-4-c(2) and Appendix C-VI.**

II. Definitions

Transgenic animal: any whole vertebrate animal, in which the animal's genome has been altered by stable introduction of recombinant or synthetic DNA into the germ-line (production of transgenic, knock-out, knock-in animals).

r/sNA: recombinant or synthetic nucleic acid molecules

III. Responsibilities

- All Dartmouth College Principal Investigators (PIs) must comply with the *NIH Guidelines* regardless of funding source. Failure to follow *NIH Guidelines* can result in the suspension, limitation, or termination of NIH funds for all recombinant or synthetic DNA research at the College.



- The PI is responsible for reporting any inadvertent release of transgenic animals, improper disposal of transgenic animals or other incidents in the laboratory or vivaria to the Biosafety Officer, who shall report them to the IBC, Attending Veterinarian, and to the NIH, if deemed necessary. Please see below for reporting procedures.
- The PI is responsible for training students, teaching assistants, and lab staff about the policies and procedures for transgenic animal handling and appropriate carcass disposal.

IV. Procedures

a. Approvals

- Research (internal and extramural funding) activities that are conducted with the goal of producing transgenic animals by stable introduction of r/sNA into the germline, as described in the *NIH Guidelines*, must be reviewed and approved by the Institutional Biosafety Committee (IBC) and the Institutional Animal Care and Use Committee (IACUC). Methods for transgenic animal production include, but are not limited to DNA microinjection, retrovirus-mediated gene transfer and embryonic stem cell mediated gene transfer.
- Experiments involving breeding of certain BSL1 transgenic rodents are Exempt under the *NIH Guidelines*.
- Experiments involving the purchase or transfer of BSL1 transgenic rodents are Exempt under the *NIH Guidelines*.
- Exempt animals still need to be disposed of as outlined below.

b. Animal Facility Security

Animal facilities housing transgenic animals are secured in accordance with *NIH Guidelines*. Containment facilities are locked and access is limited to research personnel and animal caretakers. Animal facilities are monitored on a regular basis by the Attending Veterinarian and veterinary staff, animal caretakers, research personnel, and are inspected semi-annually by the IACUC. All facilities are maintained to minimize the possibility of theft, escape or accidental release of animals.

c. Physical and Biological Containment Levels

The containment levels required for research involving r/sNA in animals is based on the classification of experiments described in Section III of the *NIH Guidelines*. Four containment levels are established in Appendix G of the *NIH Guidelines* for physical containment of small animals: Animal Biosafety Levels 1-4 (ABSL 1-4). Research requiring ABSL-3 or -4 containment is not permitted in Dartmouth research facilities.



i. NIH Section III-E-3: “Experiments Involving Transgenic Rodents (ABSL1)”:

This section covers experiments involving the generation of rodents in which the animal's genome has been altered by stable introduction of recombinant or synthetic nucleic acid molecules, or nucleic acids derived therefrom, into the germ-line (transgenic rodents). Only experiments that require ABSL1 containment are covered under this section; experiments that require ABSL2 containment are covered under Section III-D-4, “Experiments Involving Whole Animals”.

ii. Section III-D-4: “Experiments Involving Whole Transgenic Animals (ABSL2)”:

This section covers experiments involving whole animals in which the animal's genome has been altered by stable introduction of recombinant or synthetic nucleic acid molecules, or nucleic acids derived therefrom, into the germ-line (transgenic animals) and experiments involving viable r/sNA-modified microorganisms tested on whole animals. For the latter, other than viruses that are only vertically transmitted, the experiments may not be conducted at ABSL-1 containment. A minimum containment of ABSL-2 is required.

Please refer to the NIH Guidelines Section III-D-4 for more details.

d. Disposal of Transgenic Animals

All animal carcasses are disposed of according to established CCMR and Environmental Health & Safety policies and SOPs. Animal carcasses and/or tissues are collected for commercial incineration. This applies to confirmed transgenic animals, potentially transgenic animals, “no-takes” in the production of transgenic animals, and progeny of transgenic animals.

V. Reporting an Environmental Release

- Notify the Biological Safety Officer (BSO) within 24 hrs via email or phone.
- The BSO will investigate the incident and consult with the IBC Chair and EHS Director.
- If it is determined that the incident involves r/sNA molecules, the BSO will submit an incident report to the NIH Office of Biotechnology Activities within 30 days.

VI. References

[NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules](#) (2016). Department of Health & Human Services: 78 FR 66751.