Program Description

Animal Care and Use Program

National Institute on Drug Abuse (NIDA)

Intramural Research Program

000648

National Institutes of Health (NIH)

Redacted by agreement

Baltimore, Maryland 21224

AUGUST 2018

For

AAALAC International

Table of Contents

Appendices

Program Description

Instructions for Completing and Submitting the Program Description for the Institutional Animal Care and Use Program

Section 1. Introduction

A. State the name of the program unit and, if applicable, its parent organization. List all organizations (schools, centers, etc.) included within the program unit.

National Institute on Drug Abuse (NIDA) - Intramural Research Program (IRP)
National Institutes of Health (NIH)
Public Health Service (PHS)
Department of Health and Human Services (DHHS)

B. Give a brief overview of the institution, its purpose and how the animal care and use program relates to the mission of the institution.

The NIDA IRP supports an intramural, multidisciplinary program which includes basic, preclinical, and clinical research to understand the nature of drug addiction and related neurological diseases. This research includes investigating the actions of drugs of abuse, and identifying and characterizing the mechanisms responsible for the acquisition, maintenance, and elimination of drug-taking behavior and its consequences. The animal care and use program is a vital part of this research, which ranges from basic studies of the toxicity and abuse liability of new drugs to the safety of potential new medications for drug abuse treatment. The program includes the breeding of genetically engineered mice and rats as tools for these research endeavors.

C. Note that AAALAC International's three primary standards are *the Guide for the Care and Use of Laboratory Animals* (*Guide*), NRC, 2011; the *Guide for the Care and Use of Agricultural Animals in Research and Teaching* (*Ag Guide*), FASS, 2010, and the European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes, Council of Europe (ETS 123). Other regulations and guidelines used (U.S. Department of Agriculture (USDA), Public Health Service (PHS) Policy, Good Laboratory Practice (GLP), Canadian Council on Animal Care (CCAC), etc.) may also apply. Describe which of the three primary standards and other regulations and guidelines are used as standards for the institutional animal care and use program and how they are applied. For example, an academic institution in the United States with an Office of Laboratory Animal Welfare (OLAW) Assurance may use the standards of the *Guide* and PHS Policy for all animals, the Animal Welfare Act regulations for covered species, and the *Ag Guide* for agricultural animals used in agricultural research and teaching (see also *Guide*, pp. 32-33). In the European Union, the standards applied might be the

Guide, ETS 123, Directive 2010/63, and any country-specific regulations.

The NIDA IRP is included in the NIH's Intramural Research Program assurance D16-00602 (A4149-01) on file with the Office of Laboratory Animal Welfare (OLAW) and uses the standards of the Guide for the Care and Use of Laboratory Animals (Guide), NRC, 2011, and PHS Policy on Humane Care and Use of Laboratory Animals for all animals, the Animal Welfare Act regulations, and USDA Animal Care Policies for covered species. When applicable, the Guide for the Care and Use of Agricultural Animals in Research and Teaching (Ag Guide), FASS 2010, is used.

The NIH Animal Research Advisory Committee (ARAC) advises the Institutional Official (IO) regarding interpretation of regulations and policy and develops policies and guidelines to establish best practices, that, when applied, assure consistency of practice among all NIH intramural animal programs. These NIH intramural policies and guidelines can be accessed through the OACU website. A list of ARAC policies and guidelines is provided in the Appendices.

The NIDA ACUC also implements NIDA-specific policies, guidelines, and standard operating procedures. A list of NIDA-specific policies and guidelines is provided in Appendices.

D. Describe the organization and include an accurate, current, and detailed organizational chart or charts (see **Appendix 4**) detailing the lines of authority from the Institutional Official to the Attending Veterinarian, the Institutional Animal Care and Use Committee/Oversight Body (IACUC/OB), and the personnel providing animal care. Please include the title, name (*Note:* For individuals whose information is publicly available, provide the titles and names; for individuals whose information is not publicly available, you may provide titles only.), and degree (if applicable) of each individual at the level of supervisor or above. Names of animal care staff below the title of supervisor need not be included, but the titles and number of animal care personnel under each supervisor should be included. If animal care responsibility is administratively decentralized, including the management of satellite housing areas/locations, the organizational chart or charts must include all animal care programs, indicating the relationship between each administrative unit and personnel, the Attending Veterinarian, and the Institutional Official.

- Dr. Michael Gottesman, NIH Deputy Director for Intramural Research, is the Institutional Official (IO) and has delegated authority and responsibility from the NIH Director and CEO.
- Redacted by agreement
- Alex Hoffman, Ph.D. serves as the ACUC Chair.

- Kyle Stump, D.V.M. the Animal Program Director (APD) and Attending Veterinarian, receives delegated program authority from the SD.
- Redacted by agreement is supervised by Dr. Stump.
- Redacted by agreement is supervised by Redacted by agreement Redacted by agreement
- Redacted by agreement supervises the SciTech Services, Inc., contract which provides animal care and technical services. Dr. Stump is the Contracting Officer's Representative for the SciTech Services contract.
- The Satellite Animal Housing area Redacted by agreement Redacted by agreement is supervised by the laboratory's Redacted by agreement Redacted by agreement

An organizational chart, including lines of authority and supervisory staff, with the titles and number of animal care personnel under each supervisor, is provided in Appendix 4.

E. Identify the key institutional representatives (including, but not limited to, the Institutional Official; IACUC/OB Chairperson; Attending Veterinarian; animal program manager; individual(s) providing biosafety, chemical hazard, and radiation safety oversight; etc.); and individuals anticipated to participate in the site visit.

NIH Deputy Director for Intramural Research (Institutional Official)	Michael Gottesman, M.D., Ph.D.
Redacted by agreement	
ACUC Chair	Alex Hoffman, Ph.D.
Animal Program Director (AV)	Kyle Stump, D.V.M., DACLAM
Redacted by agreement	

- Biosafety, chemical hazard, and radiation safety oversight is provided by the centralized NIH Office of Research Services (ORS)/Division of Health and Safety (DOHS) and Office of Research Facilities (ORF).

F. Briefly describe the major types of research, testing, and teaching programs involving animals and note the approximate number of principal investigators and protocols involving the use of animals. As mentioned in the instructions, please complete **Appendix 5** (Animal Usage) or provide the information requested in a similar format as an Appendix.

The NIDA intramural program consists of approximately 61 principal investigators (PIs) using animals and 142 active, approved animal study proposals (ASPs) distributed across scientifically focused organizational units (Branches). Sections and units within each Branch are also defined by scientific aims. In addition, Core facilities are available to support collaborative research from multiple investigators.

Office of Scientific Director (OSD)

The **Genetic Engineering and Viral Vector Core** provides molecular biology services to develop and produce genetic tools capable of modulating and monitoring molecules, cells and circuits within the nervous system. The **Optogenetics and Transgenic Technology Core** (OTTC) generates and characterizes transgenic rats for use by NIDA and other researchers of the molecular and cellular functions of neurons in cognition, behavior and disease.

The **Electron Microscopy (EM) Core** and the **Histology Core** provide sectioning, immunostaining and counterstaining for, respectively, EM imaging and whole-brain serial tomography, and imaging of cleared sample using light-sheet microscopy.

The **Ex Vivo Electrophysiology Core** examines functional circuits and the electrical properties of neuronal cells using existing and emerging neurophysiological techniques to support the objectives of individual projects.

The **Structural Biology Core** provides techniques in mass spectrometry to determine the presence, distribution, and amounts of molecules in biological samples.

The OSD also includes:

The **Animal Care Section** (3 PIs, 4 ASPs) – provides veterinary care and animal husbandry, including centralized mouse and rat breeding services, and basic biomethodology training to support NIDA investigators.

The **Designer Drug Research Unit** (3 PIs, 6 ASPs) collects, analyzes and disseminates the most up-to-date information about the pharmacology and toxicology of newly-emerging abused drugs, known as new psychoactive substances (NPS). Methods of investigation using animals include in vivo microdialysis, biotelemetry, pharmacokinetic analysis and behavioral assays.

Cellular Neurobiology Branch

The **Behavioral Neurophysiology Neuroscience Section** (2 PIs, 4 ASPs) focuses on the neural circuits mediating associative learning and decision-making, and how alterations in those circuits contribute to maladaptive behaviors in neuropsychiatric disorders such as addiction. The laboratory uses established and novel behavioral approaches combined with techniques ranging from single-unit recording to fast scan cyclic voltammetry and cell-specific modulation of neuronal circuitry.

The **Synaptic Plasticity Section** (5 PIs, 9 ASPs) investigates drug-induced neuroadaptations in excitatory transmission and other neuronal intrinsic properties within key reward circuits and uses optogenetic technology to identify the role of specific circuits in reward-learning behaviors.

The **Electrophysiology Research Section** (4 PIs, 8 ASPs) studies the mechanisms of action of abused drugs on synaptic neuronal circuitry using voltammetry, patch clamp electrophysiology, and confocal microscopy with brain slices.

The **Neuronal Circuits and Behavior Unit** (1 PI, 6 ASPs) combines optogenetic, electrophysiological, in vivo imagine, and behavioral techniques to elucidate the neuronal basis of goal-directed behaviors, such as feeding, and to determine how these behaviors are disrupted in eating disorders and drug addiction. Experiments involve measurement and manipulation of genetically-defined cell types during behavior to identify key elements of the feeding-reward circuits and combine behavioral and cell type analysis to determine how circuits dysfunction in addiction.

Molecular Neuropsychiatry Branch

The **Molecular Neuropsychiatry Section** (3 PIs, 5 ASPs) investigates cellular and molecular mechanisms of neurodegeneration and regeneration focusing on the role of oxidative stress, and mitochondrial pathways in the pathogenesis of psychostimulant toxicity and methamphetamine's role in epigenetic changes in gene expression and cognitive dysfunctions in the rodent brain.

Integrative Neuroscience Branch

The **Behavioral Neuroscience Section** (no currently active ASPs) projects include studies focused on the mesolimbic and nigrostriatal dopamine systems and their glutamatergic and cholinergic inputs in the characterization of brain circuitry associated with drug reward and reward prediction. Studies include an examination of neuroadaptations in reward circuitry resulting from drug experience and stress.

The **Cellular Pathobiology Section** (3 PI, 6 ASPs) studies the cellular biological processes underlying addiction and related pathobiological disorders by examining signaling processes, particularly through sigma receptors, through which drugs of abuse alter neuronal structure and function.

The **Molecular Mechanisms of Cellular Stress and Inflammation unit** (2 PIs, 12 ASPs) studies the role of endoplasmic reticulum stress and inflammation in neuronal dysfunction caused by substance abuse or neurodegenerative diseases, examines the influence of drugs of abuse on microglial activation, and develops genetic and pharmacological tools to monitor and modulate ER calcium.

The **Neurobiology of Addiction Section** (5 PIs, 12 ASPs) investigates the behavioral and neural consequences of extended access to drugs of abuse and evaluates potential pharmacotherapies to treat drug addiction. The major goal is to understand the brain regions and networks that modulate compulsive drug taking in rodents using behavioral, pharmacological, neuroanatomical, and molecular techniques.

The **Neuronal Networks Section** (1 PI, 5 ASPs) applies anatomical, molecular, cell biological and electrophysiological approaches to investigate interactions between the stress and reward systems and the cellular composition and neuronal connectivity of the ventral tegmental area and associated pathways which are central to the neurobiology of addiction.

Behavioral Neuroscience Branch

The **Cell Biology of Trafficking Unit** (4 PIs, 6 ASPs) develops miniScope imaging system for in vivo deep brain calcium imaging of neuronal activity and closed-loop optical recording/stimulation device for real-time feedback control of neuronal activity in freely moving mice. In addition, research conducted in the unit applies two-photon volume imaging technique for in vivo deep brain calcium imaging of neuronal activity in head-fixed mice.

The **Neurobiology of Relapse Section** (6 PIs, 9 ASPs) characterizes the behavioral and neurobiological mechanisms of drug reward and relapse to drug use, using animal models that examine incubation of drug craving after forced or voluntary abstinence, context-induced relapse to drug seeking after punishment or extinction, and aggression addiction and relapse. The section also performs physiological analysis of motivated behavior using single-unit recording, iontophoresis, EEG/EMG, brain thermorecording, and glutamate electrochemistry in studies of the neurophysiological and neurochemical mechanisms of action of abused drugs and alterations in blood-brain barrier in normal brain functions and during drug intoxication.

The **Neurocircuitry of Motivation Section** (3 PIs, 3 ASPs) studies brain circuits and organization that regulate motivation and mood, using rodent models. Optogenetic and chemogenetic technologies, which enable cell specific, selective manipulations of neural populations, are used in combination with behavioral assays to examine whether the stimulation or inhibition of certain populations of neurons or circuits from one neural population to another produces positive or negative motivational and mood state. Additionally, intracranial self-administration procedures, where rats learn to respond for small injections of chemicals, including drugs, into discrete brain regions, are used to understand the action sites of many abused drugs for positive mood effects, and forebrain-brainstem interactions are detected by multi regional recordings of units and local field potentials in motivation and reward.

Neuronal Ensembles in Addiction Section (2 PIs, 13 ASPs) manipulate specific neuronal ensembles through the c-fos promoter to turn on different transgenes in transgenic rats, assess ensemble role in drug-related memories, and examine synaptic alterations in novel c-fos-GFP transgenic rats.

The **Preclinical Pharmacology Section** (2 PIs, 4 ASPs) employs multimodal imaging and in-vivo electrophysiological approaches in animal models of drug use to better understand the systems neuroscience of addiction and its cognitive consequences. Ongoing projects include determining the neural underpinnings of attentional bias to drug cues using high density multisite in-vivo electrophysiological recording and investigating the relationship between cognitive changes and alterations in brain structure following chronic cocaine self-

administration. Additional work focuses supporting medications development for nicotine and opiate dependence with clinically relevant animal models, and conducting preclinical investigations of the neurobiological consequences of transcranial magnetic stimulation to better understand its potential use in treating addiction.

Molecular Targets and Medications Discovery Branch

The **Integrative Neurobiology Section** (1 PI, 1 ASP) projects include functional and pharmacological significance of neurotransmitter receptor heteromers with in vivo approaches and molecular interactions involved in the quaternary structure and function of neurotransmitter receptor heteromers (cellular and molecular approaches).

The **Neuropsychopharmacology Section** (1 PI, 2 ASPs) studies neural mechanisms underlying the initiation of, maintenance of, and relapse to drug-taking behavior and the development of psychopharmacological interventions to alter these aspects of drug-taking. Pharmacological interventions include compounds acting via endocannabinoids, dopamine D3 receptors, metabotropic glutamate receptors, GABA-B receptors, and dopamine transporter. Projects also include mechanism-based medication development for treatment of addiction, optogenetic investigation of brain neural circuits modulating reward and aversion, development of magnetogenetic techniques to control drug-taking and drug-seeking behavior, development of nanomedicine and brain-target drug delivery techniques for treatment of addiction

The **Medicinal Chemistry Section** (1 PIs, 6 ASPs) designs and synthesizes novel ligands to study the role of central dopaminergic (D2-family) and metabotropic glutamatergic (mGluR5) receptors and monoamine transporters in cocaine abuse. Highly selective compounds are designed and synthesized for characterization of these molecular targets through the development of structure-activity relationships and molecular modeling.

The **Computational Chemistry Unit** (1 PI, 2 ASPs) conducts mechanistic studies of membrane proteins, such as G-protein coupled receptors and secondary-active transporters, with computational approaches including bioinformatics, molecular modeling and simulations.

Neuroimaging Branch

The **Magnetic Resonance Imaging and Spectroscopy Section** contains two units:
The Preclinical (Translational) Imaging Unit (5 PIs, 11 ASPs) which uses functional magnetic resonance imaging and microPET to study the neurochemical, neuroanatomical, and physiological substrates and mechanisms that mediate the behavioral, psychological, and physiological effects of abused drugs.
The **Biobehavioral Imaging and Molecular Neuropsychopharmacology Unit** (1 PI, 5 ASPs) develops "biobehavioral and molecular imaging" methods and technologies using state-of-the-art imaging paradigms to interrogate systems- and cellular-level molecular neurobiology in freely-moving animals and integrating neuromodulatory technologies and positron emission tomography (PET) and investigates the involvement of essential metals (e.g. zinc, iron, copper) on brain neurobiological mechanisms relevant to substance abuse

and addiction vulnerability and the interactions between peripheral and brain energy metabolic mechanisms in regulating reinforcement and behaviors relevant to substance abuse and addiction vulnerability.

G. Note the source(s) of research funding (grants, contracts, etc.) involving the use of animals.

The NIDA animal research program, as a Federal entity, is supported by federal funds and NIH administered Cooperative Research and Development Agreement (CRADA) funding.

H. List other units (divisions, institutes, areas, departments, colleges, etc.) of your organization that house and/or use animals that are not included in this Description. If any of these are contiguous, physically or operationally (e.g., same IACUC/OB, same animal care staff), with the applicant unit, describe the association. Explain why such units are not part of this program application.
Note: Questions regarding this section should be forwarded to the AAALAC Office.

No NIDA IRP-sponsored animals are housed outside of the Redacted by agreement Buildings with the exception that nonhuman primates may be housed during initial quarantine at the AAALAC accredited Redacted by agreement

I. Contract Facilities: If the institution contracts for animal care facilities or services for animals owned by the institution, the contractor and its AAALAC International accreditation status must be identified. If a contractor's animal care and use program is not accredited by AAALAC International, a brief description, following this Program Description outline, of the relevant contractor's programs and facilities must be provided. In addition, the species and approximate average number of animals housed in the contract facilities and the approximate distance between the institution's animal facility and the contract facility must be noted. Incorporation of the contractor program into the site visit schedule will be discussed with institutional representatives. If the institution does not contract for animal care facilities or services, so note.

NIDA uses no contract facilities for the animal care and use program. SciTech Services, Inc., provides contract animal care and research support for the NIDA animal care and use program in the NIDA Redacted by agreement buildings.

J. Note other relevant background that will assist reviewers of this report.

None is necessary.

Section 2. Description

I. Animal Care and Use Program

A. Program Management

1. Program Management Responsibility [*Guide*, pp. 13-15]

a. The Institutional Official [*Guide* pp. 13-14]
Describe how program needs are clearly and regularly communicated to the Institutional Official by the Attending Veterinarian, IACUC/OB, and others associated with the program.

Dr. Alex Hoffman, ACUC Chair, communicates directly with the institutional official, Dr. Michael Gottesman, NIH Deputy Director for Intramural Research. Dr. Hoffman is a member of the NIH Animal Research Advisory Committee (ARAC), a committee comprised of the NIH ACUC chairs and others that advises Dr. Gottesman on animal use policy within the Intramural Research Program. ARAC is Redacted by agreement Dr. Hoffman also reports to Redacted by agreement who communicates frequently with Dr. Gottesman in meetings twice a month of the NIH Scientific Directors. The OLAW semiannual report of research facilities and the USDA Annual report are submitted, respectively, by Dr. Hoffman and Redacted by agreement to Dr. Gottesman through the NIH Office of Animal Care and Use (OACU).

Dr. Kyle Stump, Animal Program Director (APD) and Attending Veterinarian (AV) reports directly to Redacted by agreement and participates along with senior NIDA scientists in the monthly SD's Faculty Meeting to discuss the NIDA research program and animal program needs. Animal Program issues are communicated regularly to the ACUC Chair in monthly ACUC meetings, monthly Animal Program meetings, and by frequent direct communication.

Redacted by agreement

b. Role of the Attending Veterinarian [*Guide*, p. 14]

i. Describe the institutional arrangement for providing adequate veterinary care. Although individual name(s) and qualifications will be described below, identify by title the veterinarian(s) responsible for the veterinary care program, including:

- a list of responsibilities
- a description of the veterinarian's involvement in monitoring the care and use of laboratory animals
- the percentage of time devoted to supporting the animal care and use program of the institution if full-time; or the frequency and duration of visits if employed part-time or as a consultant.
  Note: If preferred, this information may be provided in a Table or additional Appendix.

The Animal Program Director (APD) and Facility Veterinarian are full time veterinarians and devote 100% of their time to support to the animal care and use program.

The Animal Program Director reports to and receives delegated program authority from the Scientific Director (from the Institutional Official) for all activities involving animals and is responsible for ensuring compliance with all applicable regulations, guidelines and policies (as described above, Section 1.c.). APD responsibilities include:

- Serving as the Attending Veterinarian member of the ACUC,
- Reporting animal care and use issues to the ACUC, OACU, and the Scientific Director
- Supervising the Facility Veterinarian, and
- Overseeing the SciTech Services contract which provides animal care and support services for the Animal Care Program.

The Facility Veterinarian reports to the APD and has delegated program authority from the APD for all activities involving animals. The Facility Veterinarian is responsible for:

- Providing adequate veterinary care to all animals,
- Ensuring compliance of day-to-day animal facility operations, such as animal health care, husbandry and provision of supplies and equipment to meet programmatic and regulatory requirements,
- Ensuring all animal care personnel demonstrate acceptable skill in assigned duties and in performing techniques,
- Working with PIs and research staff to develop refinements in procedures and/or additions to animal activities,
- Reporting any issue of non-compliance to the APD and the ACUC,
- Serving as an alternate member of the ACUC in the absence of the APD.

ii. List others (e.g., Principal Investigators, veterinarians serving as Principal Investigators, veterinary faculty/staff, technical staff, farm managers) who have a direct role in the provision of veterinary care and describe their responsibilities. The Organizational Chart(s) provided in Appendix 4 must depict the reporting relationship between these individuals and the Attending Veterinarian.
Note: If preferred, this information may be provided in a Table or additional Appendix.

The SciTech Services contract provides one Animal Health Technician who performs NHP anesthesia support and postsurgical care and participates in the preventative and clinical veterinary care program.

The SciTech Services contract also provides a rotating roster of on-call laboratory animal veterinarians, who have completed on site orientation and are experienced with care of NIDA species, to provide emergency veterinary care in the absence of the APD or Facility Veterinarian.

c. Interinstitutional Collaborations [*Guide*, p. 15]
Describe processes for assigning animal care and use responsibility, animal ownership and IACUC/OB oversight responsibilities at off-site locations for interinstitutional collaborations.

Only NIDA-owned animals are housed at the [Redacted by agreement] Buildings, and no NIDA-sponsored animals are housed outside of [Redacted by agreement] Buildings with the exception that nonhuman primates may be housed during entry quarantine at the AAALAC accredited [Redacted by agreement]

NIH Policy Manual 3044-1 requires that entry quarantine for NHPs be performed at the NIH Animal Center. The Animal Center program assumes full responsibility for animal care during quarantine and establishes a written agreement regarding veterinary care and animal disposition with the PI of the NIDA ACUC approved ASP. The NIDA ACUC extends their semiannual program review and facilities inspections to include the NIH Animal Center when NIDA owned animals are present.

2. Personnel Management

a. Training, Education, and Continuing Educational Opportunities
Describe how the IACUC/OB provides oversight and evaluates the effectiveness of training programs and the assessment of personnel competencies. Describe how training is documented.
Note: Do not include details about the training program, which should be described in the following sections.

The ACUC provides oversight of the Animal Care and Use Training Program and ensures the program's effectiveness by:

- Tracking PI and Animal Users completion of NIH and NIDA required training (described below).
- Ensuring new ACUC members complete: "Animal Care and Use Committee Member Training: Defining the Challenge of ACUC Membership" provided by the OACU.
- Reviewing PI competency determination for each individual and the procedure(s) involving animals for which they are authorized to perform in ASP Section A. Administrative Data and in Section G.2. Survival Surgery.
- Semi-annual inspection of the facilities including animal laboratory and surgical areas and review of the animal care and use program.
- Reviewing, monthly, at least one, active ASPs with their PIs and authorized animal users and observe performance of select ASP animal procedures as part of the ACUC Protocol Adherence Program (PAV). To ensure that individuals who work with animals are experienced and appropriately trained and qualified to perform technical, hands-on procedures, PAV review includes review of all aspects of animal welfare defined in the ASP, including body weight and postoperative records; inspection of anesthetic, analgesic, and euthanasia drug inventory; evaluation of PPE and surgical instrument sterilization practices; and inspection of housed animals' general health, environmental enrichment, and social housing status. PAV program activities are communicated through the ACUC chair to PIs and the ACUC.
- Assessing effectiveness and personal competency by evaluating causes of noncompliance, addressing timely reports from investigators of adverse or unanticipated events and addressing other concerns involving the care and use of animals at the institution,
- Awareness of monitoring by a multidisciplinary team of individuals, e.g. daily observation of animals by trained animal care personnel and communication to the veterinary staff for follow up, facility monitoring by facility management personnel, post-operative care by trained personnel, evaluation of outcomes of animal procedures by investigators and staff, hands-on training in animal procedures, and appropriate reporting of incidents involving occupational health and safety.
- Identifying training needs for staff who work with laboratory animals and communicate those needs, as appropriate, to the APD, SD, or OACU.

i. Veterinary and Other Professional Staff [*Guide*, pp. 15-16]

For the Attending Veterinarian and other individuals having a direct role in providing veterinary medical care (veterinarians, other professional staff listed above, private practitioners, etc.), provide: name, credentials (including degrees), and a description of their qualifications, training, and continuing education opportunities.

Note: Please do not provide curriculum vitae of personnel; if preferred, this information may be presented in a Table or additional Appendix.

Kyle Stump, DVM, DACLAM / Animal Program Director and Attending Veterinarian	Michigan State University, DVM 1979; Johns Hopkins University, Laboratory Animal Medicine Residency 1984-87, Board certified DACLAM 1990, Licensed in State of MD. CE: DACLAM Forum, PRIM&R, OLAW, AAALAC, and AALAS webinars, research and professional seminars

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ii. Animal Care Personnel [*Guide*, p. 16]

1) Indicate the number of animal care personnel.

Animal care program staff totals 34 which are delineated by category in Appendix 4 Organizational Chart

2) Summarize their training, certification level and type, experience, and continuing education opportunities provided.
Note: If preferred, this information may be provided in a Table or additional Appendix.

Certifications	Number of staff	Animal Care Experience	Number of Staff
AALAS ALAT	11	0-2 years	6
AALAS LAT	1	3-5 years	7
AALAS LATg	6	5-10 years	1
AALAS ILAM	2	>10 years	5
AALAS CMAR	1		
RVT	1		

The animal care staff must meet all training requirements of the contractor's comprehensive training plan, developed in response to the contract Statement of Work and approved by the APD.

Corporate staff provide occupational health and safety compliance (OHSC) training in New Employee Orientation to all contract animal care and support staff at time of initial employment. A sequenced curriculum of OHSC topics are delivered monthly to animal care staff.

Personnel whose duties include entering rooms or areas that contain NHPs receive NHP/Cercopithecine Herpes Type 1 virus awareness training in a "Working Safely with Nonhuman Primates" presentation (Content is described below at iii. The Research Team), and complete post course examination.

Basic Skill Competency Training instructs animal care staff in technical and non-technical job skills to ensure performance in accordance with established policies, regulations, Standard Operating Procedures, and individualized ASP requirements. A matrix of tasks with requisite skills is developed for each job position. As each employee demonstrates sufficient competency and knowledge of the skill, a record is kept to document achievement after completing training.

The contract requires that animal caretakers be ALAT certified within one year of eligibility. Continuing education opportunities are provided to foster personal and professional development. Animal care staff is provided web based training and access to additional materials (AALAS Learning Library) to prepare for the AALAS Assistant Laboratory Animal Technician (ALAT) certification, a minimal required level for all animal care staff, and the more advanced Laboratory Animal Technician (LAT) and Laboratory Animal Technologist (LATg) tiers. Corporate staff provide two annual, personal/professional development workshops to animal care staff in supervisory and leadership roles, and the animal care staff attend the local AALAS NCAB District meetings and workshops. Selected staff may also attend the National AALAS meeting and courses offered by Charles Rivers Laboratories and Jackson Laboratories.

iii. The Research Team [*Guide*, pp. 16-17; 115-116; 122; 124]

1) Describe the general mechanisms by which the institution or IACUC/OB ensures that research personnel have the necessary knowledge and expertise in the animal procedures proposed and the species used.

The ACUC requires Principal Investigators and all animal research personnel including technicians, postdoctoral fellows, students, and visiting/guest scientists to complete the role-appropriate NIH web-based training courses

entitled "Using Animals in NIH Intramural Research" for and animal users. The ACUC Administrator verifies certification of training of all individuals listed on the ASP prior to approval of the ASP and of personnel additions to the ASP. Refresher training for both courses is required every 3 years.

In the ASP, the ACUC requires Principal Investigators to correlate named research staff with proposed animal procedures and to provide assurance that the participating staff are fully qualified or will be provided training to achieve competency before performing animal procedures independently.

- All animal research staff must certify that they have read and are familiar with the ASP on which they will be working.
- Research staff also complete a checklist description of prior experience conducting animal procedures including specific surgical procedures, euthanasia methods, and behavioral tests.
- For individuals working with conscious nonhuman primates, the Principal Investigator must provide written certification of demonstration of procedural competency.
- The ACUC provides PIs with recommended performance standards in NIDA Rat and Mouse SOPs to apply in assessing proficiency for some common animal procedures.

a) Briefly describe the content of any required training.

The NIH course "Using Animals in NIH Intramural Research: Guidelines for Animal Users" consists of four modules: 1) The *Animal Care and Use Policies* module reviews the laws, regulations and policies relevant to humane methods for animal care at NIH and also discusses relevant ethical and scientific issues related to the proper care and use of research animals. 2) The *Occupational Health and Safety* module reviews the function and requirements of the NIH occupational health program (Animal Exposure Program), appropriate Personal Protective Equipment (PPE) for working with various animal species to ensure personal and animal safety, and required security precautions and the rationale for those precautions 3) The *Animal Health and Well-Being* module describes the procedures, conditions and environmental factors that can affect the health and well-being of laboratory animals and potentially confound research results, review the procedures to follow if an animal health problem is observed, and introduces select animal husbandry topics such as caging types and proper methods of transporting various species. 4) The *Animal Care and Use Procedures* module describes, in detail, the range of procedures that the Animal User may need to perform on laboratory animals including general principles of restraint, reviews the general principles for giving injections and taking samples, administering anesthetics and analgesics, describes the difference in requirements for performing survival surgery on

rodents and larger species (e.g. non-human primates), and reviews the general principles for humanely euthanizing animals.

The NIH course "Using Animals in NIH Intramural Research: Guidelines for Principal Investigators" contains four modules: 1) *Animal Care and Use Policies* and 2) *Occupational Health and Safety*, are identical to those included in the "Using Animals in Intramural Research" course, described above. 3) The *Animal Study Proposal Composition* module reviews the general information needed to successfully complete each section of the Animal Study Proposal (ASP) and responsibilities, as Principal Investigator, in the ASP process, and 4) the *Animal Study Proposal Maintenance* module covers the Principal Investigator's (PI) responsibilities for addressing training and experience requirements for themselves and the Animal Users listed on their Animal Study Proposals (ASP) as well as how to address ASP compliance and maintain ASP currency through periodic reviews and modifications.

The NIH Course, "Using Animals in NIH Intramural Research: Refresher" is a scenario based overview of the regulations governing animal care and use in NIH research and describes the structure of the NIH animal program, focusing on the organizations and individuals with oversight responsibility, and the specific responsibilities of the Principal Investigator and the Animal User in the oversight and conduct of research using animals at NIH.

The Facility Veterinarian and ACUC Administrator presents a NIDA "**New Animal Users Orientation**" course to all new animal users, and the Facility Veterinarian also presents an interactive "**Support Personnel Orientation**" course for building engineers and craftsman that may need to enter the vivariums as a part of their daily routine work requirements or for emergency purposes. The "New Animal Users Orientation" topics are tailored to the animal research program and user and include:

- Reporting animal welfare concerns
- Accessing ACUC SOPs and Guidelines
- Accessing Veterinary Care
- Occupational Health Requirements
- Occupational risks of animal contact including allergens
- PPE requirement review
- Transporting animals within and outside of the vivarium
- Euthanasia practices and service
- Animal research regulation review
- ACUC protocol review process and ASP compliance
- Documenting SOP and ASP practices
- Registering for the Animal Program's basic biomethodology classes

A contact list of key Animal Program personnel is also provided. The presentation is followed by a tour of the vivarium with the SciTech Services contract Facility Manager.

All NIDA rodent surgeons view an on-line NINDS Aseptic Techniques Training Course which provides general training in rodent aseptic technique regarding procedure area set up, sterile instrument preparation, animal preparation, and surgeon's attire and gloving. All surgeons complete a post course mastery quiz. The Animal Program Director reviews the completed quiz with the prospective surgeons.

All NIDA animal users working with NHPs receive introductory "**Working Safely with Nonhuman Primates**" course from the FV to cover key NIH Safety regulations, video presentation & quiz, review of personal protective equipment (PPE) requirements, and location of Macaque Exposure Kits (which contain first aid supplies, valacyclovir and written first aid and reporting instructions), eyewash stations, and training resource prior to technical training with NHPs, and a post course mastery quiz. The quiz documents animal user completion of the course and review of post course examination results with the FV.

Live primate training and certification of animal users is performed by trainers in the laboratory in which the individual will be working. This training is specific to the ASP procedures used in each of the laboratories and covers restraint procedures (squeeze back, pole and tether, chair, and hand restraint), acclimation procedures, injections, drug infusion, and feeding procedures. Competency assessment is performed and documented by the PI or designee and additional training (if needed) is provided before individuals are permitted to work independently.

b) Describe the timing of training requirements relative to the commencement of work.

All animal users must complete the NIDA **New Animal Users Orientation** to obtain independent vivarium access and must complete the NIH **Using Animals in NIH Intramural Research** course prior to working with animals.

Rodent surgeons and NHP users must complete, respectively, **Aseptic Technique Training Course** and **Working Safely with Nonhuman Primates** courses prior to starting PI provided technical training.

In addition, prior to ASP approval, an Animal Assurance Training from is required to be signed by all personnel listed on the ASP. The form attests that all personnel have read the ASP and understand their responsibilities for listed procedures. The PI must also sign the assurance form to indicate

their responsibility for training personnel and assessing competency in procedures.

c) Describe continuing education opportunities offered.

All animal users within the NIDA are emailed updates regarding best practices, regulation or institutional policy changes, animal rights activities and items of general interest regarding the humane care and use of animals. In addition, the NIDA animal program maintains a secure, internal web page for easy access to operating procedures, guidance documents, staff and ASP information, shared procedure room reservation, training room reservation, training scheduling, form downloading and ACUC schedules.

The animal program uses webinars provided by OLAW, AALAS, and commercial animal vendors (e.g. Jackson laboratories and Charles River Laboratories) and posting educational information within the vivarium for NIDA staff.

2) Describe the process(es) to ensure surgical and related procedures are performed by qualified and trained personnel, including:

- who determines that personnel are qualified and trained for surgical procedures
- the roles that the Attending Veterinarian and IACUC/OB have in this determination [*Guide*, pp. 115-116]

Rodent surgeons are required to view the NINDS Aseptic Technique Training Course and complete a post course mastery quiz. The Animal Program Director (Attending Veterinarian) meets with all prospective surgeons to review the completed quiz and discuss the expectations for animal surgery and post-operative care.

The Principal Investigator determines that personnel are qualified and trained for rodent surgical procedures. Rodent surgeries are performed by research staff which have been identified and trained by the PI or PI's designee, typically experienced research technicians, to perform specific operative techniques (e.g. intracranial cannulation, intravenous catheterization). The APD and Facility Veterinarian also offer training on general surgical skills (e.g., tissue handling, suturing, hemostasis, instrument handling, and maintaining a sterile field). As described above (Section 2.a. Training, Education, and Continuing Educational Opportunities), the ACUC reviews PI competency determination for each individual and the procedure(s) involving animals for which they are authorized to perform in ASP Section A. Administrative Data and in Section G.2. Survival Surgery.

The veterinarian(s) determines that personnel are qualified and trained for NHP surgical procedure. One researcher, who has over thirty years of experience implanting chronic intravenous catheters in Squirrel monkeys and performs most Squirrel monkey catheter surgery, is training two other researchers with experience in human and rodent surgery. Although not yet initiated, the PI involved with Rhesus monkey research has 25 years of experience with surgical placement of chambers for intracranial procedures and chronic intravenous catheters in Rhesus monkeys. The Veterinary Care staff provides anesthesia and postsurgical support.

3) Describe the training and experience required to perform anesthesia. [*Guide*, p. 122]

The **"Using Animals in NIH Intramural Research"** course, NIDA ACUC Anesthesia Guidelines, and the species-specific SOPs also provide detailed information on injectable and inhalation anesthesia and proper monitoring.

The PI trains individuals in the specific anesthesia methods used in their rat and mouse studies; the Facility Veterinarian provides and oversees anesthesia for Squirrel Monkey and Rhesus Monkey procedures.

4) Describe how the proficiency of personnel conducting euthanasia is ensured (especially physical methods of euthanasia). [*Guide*, p. 124]

The veterinarian(s) performs or, for perfusion, directly oversees all euthanasia of nonhuman primates for the researchers.

The animal care staff provides a CO2 euthanasia service for rodents. Detailed instructions on carbon dioxide euthanasia are posted at each euthanasia station. All animal care staff receive hands-on instruction in carbon dioxide euthanasia and in performing the required secondary physical methods of decapitation (or, bilateral pneumothorax as an alternative) in rats and cervical dislocation in mice.

Principal investigators are responsible for training their research staff in performing the euthanasia method(s) described in the approved ASP.

- The NIDA ACUC requires use of a physical method as the primary method or, after use of a chemical method, as a secondary procedure of euthanasia.
- NIDA Rat and Mouse SOPs (Section 8) promotes the use of euthanized or deeply anesthetized rodents for the training of inexperienced research personnel and recommends performance standards to apply in assessing proficiency of trainees.

b. Occupational Health and Safety of Personnel [*Guide*, pp. 17-23]

i. Institutional Oversight [*Guide*, pp. 17-19]

1) List the institutional entities (units, departments, personnel, *etc.*) that are involved in the planning, oversight, and operation of the institutional occupational health and safety program related to animal care and use (e.g., office(s) of environmental health, institutional health services or clinics (*including contracted health services*), industrial hygienists, Institutional Biosafety Committee(s) and/or Officer(s), Radiation Safety Committee(s) and/or Officer(s).

- Include a brief description of their responsibilities and qualifications.
- If contracted services are used, also include their location (e.g. remote offices to which personnel must report).

The NIH Office of Research Services (ORS), through the NIH Division of Occupational Health and Safety (DOHS) and the NIH Division of Radiation Safety (DRS) serve as the primary operational component in developing and implementing NIH-wide safety and health programs through surveillance, consultation, training, and education, provides administrative management for the comprehensive NIH Occupational Safety and Health Management Program. The Office of Research Facilities (ORF) provides environmental health and waste management services for general, chemical, multi-hazardous, radioactive, and medical pathological wastes (MPW).

- The DOHS Safety and Occupational Health Branch(SOSB) stations three positions to provide Occupational Health and Safety needs at NIDA: Safety Manager, Occupational Safety and Health Specialist, and Chemical Hygienist. This Safety department functions to:
  - Provide occupational safety advice, guidance, and oversight of the laboratory animal research program and support to the ACUC.
  - Obtain specialized support from the DOHS Animal Program and Safety Manager.
  - Implement biological and chemical safety policies and provide oversight and support to provide services to federal employees and assist contractors when needed.
  - The NIDA Scientific Director appointed the designated Occupational Safety and Health Specialist (with Safety Manager as an alternate) to serve as a full voting member of the ACUC. The Specialist serves as a safety and health resource to the committee. The Specialist fully participates in the ASP review/ approval process. His/ her primary focus is to identify and mitigate hazards associated with the work described in the ASP. Each ASP has a section (Section K.) completed by the PI indicating all hazardous materials (infectious, radiological, chemical, recombinant DNA) that will be used in the protocol.

In addition, a protocol-driven risk assessment is performed; mitigations identified; and safety information and requirements are communicated to the research and support staff through the ASP process. Concurrence of review and approval by the Safety Specialist is required prior to final approval of the ASP.

- Occupational Medical Service (OMS) of DOHS implements the Animal Exposure Program (AEP), a mandatory program, for personnel in direct contact with live animals or with unfixed, Old World NHP animal tissues, supports the DOHS Laboratory Animal Allergy Prevention Program (LAAPP), and provides continuous oversight, including 24/7 telephonic access to an occupational health physician. On-site NIDA OMS staffing includes registered nurses to provide medical services five days a week.
- Technical Assistance Branch (TAB) of DOHS provides industrial hygienists to implement the Ethylene Oxide (EtO), Formaldehyde, and Waste Anesthetic Gas Surveillance Programs and consultative support, upon request, to contract building managers which support all forms of animal research.

The Division of Radiation Safety (DRS) assigns a Health Physicist to oversee NIDA radiation use including the execution of NIH Radiation Safety Committee approved Animal Study Proposals, and the Division of Environmental Protection of ORF provides an on-site Environmental Protection Manager.

IRP and transmural Safety Committees assist the DOHS and DRS with the organization and administration of the OSH Management Program at NIDA.

The NIDA IRP Safety and Health Committee serves as a conduit for communication between IRP employees, DOHS, and the DRS concerning occupational safety and health matters and communicates, as appropriate, recommendations, survey results and corrective actions to the IRP SD, NIDA Director, NIH Occupational Safety and Health Committee (OSHC), and the Director of the Office of Intramural Research. The Safety Specialist), appointed by the SD as a full member to the Safety and Health Committee, provides technical safety services and advice to the Committee, as well as disseminates relevant information to the Animal Program, as needed. Meeting on a quarterly basis, the Committee:

- Develops IRP specific OHS policies and procedures in coordination with the DOHS and the DRS;
- Assists in the development of prevention strategies for work related accidents resulting in personal injury, illness and/or property damage;
- Oversees handling of employee suggestions, recommendations and reports of hazardous conditions.

- Monitors performance and effectiveness of safety and health activities.
- Performs annual workplace surveys with the assistance of the DOHS to assure compliance with NIH and OSHA safety and health policies and standards; conducts quality assurance checks of biological sample inventories associated with non-registered laboratories.
- Assist the Office of Intramural Research, the DOHS and the DRS in the dissemination of safety information in the event of an emergency or on an as-needed basis.

The NIH Institutional Biosafety Committee (IBC), through the DOHS

- Reviews and approves submitted documents for work with biological materials that require registration.
- Assigns appropriate biological safety level and determines any requirements such as specific PPE, containment equipment, or research practices.
- Provides advisory support by recommending safety policies and practices regarding pathogenic organisms/vectors as well as toxins of biological origin.
- Performs functions as specified in the "NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules."

The NIH Radiation Safety Committee,

- Ensures safe use of all radioactive materials and sources of radiation throughout NIH and NIH-occupied buildings within the NIH Radiation Safety Program.
- Reviews the Radiation Safety Program at least annually to determine that all activities involving radioactive materials and sources of radiation are being conducted safely and in accordance with applicable regulations.
- Provides technical advice, assistance, and management level support to the Radiation Safety Officer in implementing the Radiation Safety Program and NIH program for maintaining radiation exposures to as low as reasonably achievable (ALARA).
- The Radiation Safety Program, executed by the Division of Radiation Safety, includes:
    - Provision and documentation of radiation safety training to individuals who work with sources of ionizing radiation and to ancillary personnel.
    - Provision of comprehensive consultation to individuals using radioactive materials.
    - Receipt, inspection, and delivery of packages containing radioactive materials.
    - Disposal of radioactive waste through a DRS contract and in coordination with the ORF Environmental Protection Manager

In addition to the institutionally provided service, contractors are required to provide an occupational health and safety program which must be, at a minimum, equivalent to the NIH OHS program for their individual employees who participate in the animal care and use program. SciTech Services, Inc., provides on-site OHS training to their animal care and use employees and contracts through Concentra, a regional urgent and occupational health care provider with a local office a short (fifteen minute) drive from NIDA, for routine occupational medical services. Contractors work closely with NIDA personnel to ensure compliance with NIH standards and report injuries and potential hazards or exposures to facility manager or contract supervisor and OMS, as needed.

2) Describe methods to identify work-related hazards and the processes used to evaluate the significance of those hazards in the context of duties and tasks. Describe both common approaches and differences, if applicable, for categories of personnel such as, but not limited to, researchers, veterinarians, husbandry staff, cage-washing staff, students, housekeeping, physical plant staff, security personnel, IACUC/OB members (including non-affiliated members), contractors, visitors, etc. [*Guide*, pp. 18-19; see also Chapters 2 and 3 in Occupational Health and Safety in the Care and Use of Research Animals, NRC 1997.].

Multiple, overlapping methods and processes are applied to identify and evaluate the significance of work place hazards in context of job duties and tasks and, as described below (in Section b. iii.2), experiment related hazards. While personnel may have different job duties, the assessment of their work space, procedures and materials can all be reviewed by the IC Safety department, other DOHS entities including OMS, facilities, and/or appropriate resources for contractors

The NIDA Safety department and NIDA OMS office identify and evaluate the work-related hazards.
The Safety department

- Reviews each hire's "Anticipated Work Activities Inventory" form, prepared with assistance of their supervisors, shortly after their arrival, and interviews each new hire to individualize exposure risk assessments.
- Coordinates support from DOHS staff, including industrial hygienists, to operate several programs:
  - Anesthetic Gas Surveillance Program
  - Ethylene Oxide Surveillance Program
  - Hearing Conservation Program
  - Hazard Communication Program (including Chemical Hygiene Plan)
  - Heat Stress Program
  - Laboratory Animal Allergy Prevention Program (LAAPP)

- Respiratory Protection Program
- Ergonomic Program

The NIDA OMS (or contractor provided equivalent) provides personalized identification and evaluation of work related hazards at enrollment in the Animal Exposure Program and revised, as needed, throughout the employees' tour of duty. OMS communicate directly with the Safety Department to review injury/illness reports and contractor OSHA 200 logs to identify emerging workplace hazards. Please refer to Section 2.b. ii.1 (Medical Evaluation and Preventive Medicine for Personnel), further below, for more information regarding the Occupational Health Program.

Workplace Audit and Survey process

Ongoing assessment of research at NIH is conducted during periodic workplace audits and surveys conducted by the Safety Specialist. This includes pre-occupancy inspections by the Safety Specialist, and, for animal use areas, also by the Animal Program Director, that evaluate the workspaces prior to research beginning, as well as annual and semiannual surveys of laboratory and animal research areas. The spaces are evaluated to ensure that work in research laboratories and animal housing facilities is conducted in compliance NIH policies and industry guidance documents. Please refer to Section 2.b.i.3, immediately below, for more information on specific periodic workplace audits and surveys.

3) Describe methods and frequency of reassessing work-related hazards.

Oversight of safe work practices is provided through the conduct of audits, surveys and monitoring programs. Safety and health audits and surveys are conducted through a variety of mechanisms.

- During ACUC semi-annual "walk-throughs" of all areas in which live animals are used, a member of safety and health personnel participates on the audit teams.
  - At least twice a year, adherence to safety requirements, including those incorporated into ASPs, is monitored in animal facilities and corrections are made immediately, if possible.
  - Facility departures which may affect the safe operation of animal facilities may be identified at this time. Items requiring corrective action/remediation are recorded and tracked through resolution.
  - Compliance with the Laboratory Animal Allergen Prevention Program is monitored including use of warning signs and appropriate PPE.
- Registered Laboratories (those working with human pathogens, human or nonhuman primate blood body fluids or tissues, recombinant or synthetic nucleic acid molecules, or pathogens requiring special

permitting) are surveyed annually. Surveys are based on the assigned biosafety levels (BSLs).

- Over 100 items on 20+ safety topics are reviewed including animal-related issues, laboratory animal allergy concerns, training, PPE, housekeeping, biological safety cabinets, chemical fume hoods, cylinders, chemicals, flammables, waste management, fire hazards, etc. Infractions are documented and resolutions are tracked.
- If animals are present in these laboratories, a laboratory animal allergy warning sign is required outside of the vivarium space.
- Survey results are entered into a computerized information system (HealthRx, Lab safety module) which sends results and potential hazards to the Principal Investigator. Imminent hazards are flagged for immediate response and correction.
- PIs are responsible for correcting identified issues and recording corrective actions in the system. Corrective actions are monitored by the NIDA Safety Specialist.

• Annual surveys of non-registered laboratories are performed by the Safety Specialists during the ACUC "walk-throughs" of areas to which live animals are brought and by the NIDA Safety Committee to all other non-registered laboratories outside of the vivarium including those where only rodent tissues and fluids are brought.

• DRS assigns a health physicist to NIDA and conducts or oversees

- At least semi-annual, announced or unannounced, comprehensive surveys.
- A daily contamination survey is required each day unsealed radioactive material (RAM) is used.
- Monthly Radiation Surveys of laboratory modules are required every calendar month when unsealed forms of radioactive material have been used since the last monthly survey.

• Audits may be conducted "for cause" as well as at the request of employees; employees may anonymously request, directly to the Safety department, inspection and audit of any alleged unsafe or unhealthful workplace condition.

4) Describe institutional programs or methods used to track and evaluate safety-related workplace incidents, including injuries, exposures, accidents, etc. Include the frequency of such assessments. [*Guide*, pp. 18-19]

As part of their initial health and safety training, employees are instructed to report observed hazards, exposure incidents or near-misses to their supervisor, or any supervisory employee, and to the Safety Department and OMS. Contract employees report to the Safety department through their supervisors.

Work-related illness and injuries are reported through OMS.

- During normal duty hours, both federal and contract employees that sustain injuries and/or illnesses related to their work are eligible for the initial assessment and treatment of a work-related injury. Federal employees are instructed to report directly to OMS for initial medical triage and completion of an NIH injury report. Contract employees are instructed to report to OMS if less than 24 hours have elapsed since the injury. After that 24-hour period, the employee may report to OMS to complete an NIH injury report, but will need to obtain medical care through their employer's medical provider.
- Each injury report is assigned to the Safety and Health Specialist who is responsible for investigating the incident. They identify contributing factors (e.g., lack of PPE, training, or technique, etc.) and use the collected information to determine the root cause of the incident.

The DRS's Radiation Safety Program assigns dose(s) to individuals and issues, collects, and submits dosimeters for evaluation. DRS reports exposure finding to the individual, upon request, and investigates and reports unusual exposures to appropriate management.

The NIDA IRP Safety and Health Committee considers safety related incidents as part of their review and evaluation of the effectiveness of the IRP occupational health and safety program. The committee provides assistance with evaluation of potential any reoccurring incident investigation focus (RIIF) and recommendations in development of SOP and training programs.

The Safety Specialist and or Manager also attends monthly animal program meetings to report and discuss safety issues and accidents/injuries with the animal care program supervisory staff.

ii. Standard Working Conditions and Baseline Precautions

The following section pertains to the Occupational Health and Safety Program for all personnel associated with the animal care and use program. Specific information regarding the use of hazardous agents is included in ***subsection iii*** below.

1) Medical Evaluation and Preventive Medicine for Personnel [*Guide*, pp. 22-23]
Note: Include blank forms used for individual health assessment as **Appendix 6.**

a) Describe who (e.g., personnel assigned to job/task categories in I.A.2.b.i.2 above) receives personal medical evaluation as a component of individual risk assessment. Describe who are ***not*** included and/or exempted from personal medical evaluation. *Note:* Do not include the names of personnel.

All personnel, including trainees, fellows, and special volunteers, who have direct contact with or are involved with the direct care of live research

animals or have contact with unfixed, Old World NHP tissues receive a personal medical evaluation upon enrollment in the NIH Animal Exposure Program (AEP).

Contracts which furnish personnel who perform animal care and use procedures, building/physical plant support, security services, and housekeeping services within animal laboratories, animal housing rooms, vivarium corridors, and mechanical support for animal occupied spaces, contains a standard clause which requires meeting all regulatory and programmatic requirements for an occupational safety and health program (whose components include occupational medicine, hazard training, respirator medical clearance, fit-testing, selection, and training) that is consistent, as appropriate, with NIH policies and procedures. The contractor must provide documentation to demonstrate compliance with these specifications.

Transient visitors are not required to participate in the NIH Animal Exposure Program. A transient visitor is anyone without approved access to the animal facilities who needs to enter an animal housing or dedicated testing room, but who will not have direct contact with animals (live or dead), their viable tissues, body fluids, wastes, or caging equipment.

b) Describe provisions for allowing an individual (following completion of individual health and job related, risk assessments) to decline participation in all or part(s) of subsequently available medical and preventive medicine components of the institutional program, e.g., vaccinations, physical examinations, respiratory protection, as applicable. Provide an estimate (percentage) of personnel associated with the animal care and use program that have declined participation in the medical evaluation program.
Note: Do not include names of the personnel

When employees are enrolled in the Animal Exposure Program, the OMS nurse provides counseling regarding health concerns related to animal work, including the importance of immunizations and medical screening, if indicated. OMS reports enrollees as either "compliant" or "not compliant" with AEP medical requirements for each specific class of animals (small, large, NHP)

c) Describe provisions for assuring confidentiality of medical information.

Medical records are maintained separately by OMS, observing all standard privacy requirements. Only OMS medical staff has access to these records, and the only medical information reported to safety staff and animal care supervisors is whether or not the employee is compliant with the medical requirements required by the AEP.

d) Describe safety considerations for individuals with incidental exposure to animal care and use (e.g., contractors, personnel working in open laboratories).

Transient visitors, including ORF personnel, physical plant staff, security personnel, contractors, etc that may enter the animal facilities or have indirect contact with animals, body tissues, wastes, or living quarters, are required to wear appropriate PPE when visiting animal care and use areas. Regular work practices are reduced or, when feasible, suspended while transient visitors are present. Work with hazardous chemicals and biological materials does not take place when transient personnel are present.

Laboratory Animal Allergy Protection Program (LAAPP) warning signs are posted in all animal use laboratories outside of the vivarium and common use freight elevators used for animal transport.

e) Describe general features of the medical evaluation and preventive medicine programs, within the context of work duties, including:

- pre-employment/pre-assignment health evaluation,
- medical evaluations (including periodicity),
- diagnostic tests (e.g., for tuberculosis),
- precautions for working with potentially hazardous species (e.g., nonhuman primates, sheep, venomous species)
- immunization programs, and
- procedures for communicating health related issues.

All NIH personnel who have direct contact with or are involved in the direct care of live research animals or have contact with un-fixed, Old World NHP animal tissues or body fluids must be enrolled in the Animal Exposure Program (or contractor provided equivalent).

Pre-employment/pre-assignment health evaluation

Based on the pre-assignment evaluation, including a review of the worker's personal medical history, occupational duties, and the species of animals with which the employee may have contact, the employee is enrolled in one or more of the four sections of the AEP (small animal species, large animal species, nonhuman primates and nonhuman primate tissues).

- Appropriate vaccinations (e.g. tetanus, rubeola, etc.), if medically indicated, are provided based on medical history and laboratory testing.
- Employees who will be exposed to NHPs are evaluated for evidence of recent infection with M. tuberculosis by tuberculosis skin testing or, if vaccinated, by medical history.

- During enrollment, all workers enrolled in the AEP are entered into an OMS electronic database to support notification of the employee's supervisor and the APD indicating completion of the AEP requirements. Approved representatives are provided a read-only view of their portion of the database to confirm enrollment and active status.

Medical Evaluations (including periodicity)
The OMS medical director defines specific risk categories which require periodic personnel evaluations. The NIH OMS does not consider standard rodent users to warrant a periodic medical evaluation.

AEP participants identified as having contact with NHPs or their living spaces are recalled annually for tuberculosis testing. Workers are notified annually by email of their need to return for repeat testing for infection with M. tuberculosis. If the worker fails to return within a month of the recall notice, a second email notice is sent to the worker and copied to his or her supervisor. If the worker does not return within two weeks of the second notice, an email is sent to the NIDA APD veterinarian, notifying him/her that the worker has been removed from the AEP. The worker and his or her supervisor are copied on the removal notification.

Diagnostic tests (e.g., for tuberculosis)
Employees who are expected to have contact with nonhuman primates are evaluated during AEP enrollment by testing for protection to rubeola and prior infection with M. tuberculosis.

Precautions for working with potentially hazardous species (e.g., NHPs)
Employees who are expected to have contact with nonhuman primates are provided detailed counseling, a handout describing Macacine herpesvirus 1 (Herpes B), and a wallet card that includes a description of first aid measures, OMS contact numbers, and a description of the signs and symptoms of infection with Herpes B.

If the injury to federal or contract staff involves a possible exposure to B virus or potentially life-threatening human pathogen, OMS provides ongoing care including: laboratory testing, medications, and counseling. OMS medical providers are on-call 24 hours a day to respond to medical emergencies. Valacyclovir is included in NHP bite-scratch kits and chemoprophylaxis, if clinically indicated, is initiated on the direction of OMS medical staff.

Immunization programs
AEP enrollment evaluation includes administration of occupationally-indicated immunizations

- All animal users are offered tetanus vaccination if lacking evidence of vaccination within the preceding 10 years.
- Employees who are expected to have contact with nonhuman primates and lack protective titer or evidence of recent vaccination for rubeola are offered immunization.

Procedures for Communicating Health Related Issues.
Employees are screened during a pre-placement medical evaluation and are counseled on and provided information specifically regarding the potential for animal allergens and preventative measures.

- Enrollees are counseled regarding the need to report all injuries, first aid measures, accessing medical care after hours, allergies to laboratory animal proteins, and other suspected health hazards in the work environment.
- Enrollees are informed of the required procedures for contacting OMS if their health status changes during the course of their work, including signs and symptoms of animal allergy, as an NIH employee.
- Enrollees are provided with handouts that provide background and contact information for OMS, information on allergies to laboratory animals, Standard/Universal Precautions, and information regarding zoonoses associated with the type of animals they will handle.

Reminders are provided to employees through measures such as:

- Including health related issues in the Office of Animal Care & Use triennial Refresher course for PIs and Animal Users
- Posting animal use areas with DOHS 'animal use' signs;
- Distributing an annual OMS email notice to all AEP participants reminding them of occupational health risks associated with working with animals, the steps they can take to avoid injury, and what they should do if an accident occurs. periodic health and safety information.

f) Describe any other entities that provide medical services (e.g., emergency care, after-hours care, special medical evaluation, contracted services). Include a brief description of their credentials and/or qualifications, and how these entities remain knowledgeable about animal- or institution-related hazards and risks.

Emergency medical support for work-related injuries is available to all workers at the NIH 24/7.

- The NIDA OMS office is staffed by registered nurses during the workweek, and two OMS medical providers, including a physician at all times, are on-call through a telephonic emergency service.

- If necessary, the on-call OMS providers can contact and follow-up with on-campus medical care providers at the neighboring Johns Hopkins Bayview Emergency Room and an urgent care facility (PatientFirst) and direct employees to obtain physician care on weekends, holidays, and evenings until the OMS office is reopened. All OMS clinicians are specifically trained in the care of employees experiencing zoonotic illness or injury and receive related continuing education on an annual basis.

2) **Personnel Training Regarding Occupational Health and Safety** [*Guide*, p. 20]
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Describe general educational program(s) to inform personnel about:

- allergies,
- zoonoses,
- personal hygiene,
- physical injuries in animal facilities (e.g., noisy areas, large quantities of chemicals such as disinfectants, ergonomics) or species used (e.g., nonhuman primates, agricultural animals),
- other considerations regarding occupational health and safety.

Include in the description a summary of the topics covered, including:

- Entities responsible for providing the training
- Frequency of training or refresher training

Note: Do not include special or agent-specific training for personnel exposed to experiment-related hazardous agents; this will be provided in **Section iii.3** below.

DOHS, OMS, and animal program staff implement educational training as part of their safety programs to reduce employee exposure to hazards which are inherent to animal facilities due to their physical nature of the work involved, the requirements of facility sanitation, and the species used.

- The OMS (or contractor equivalent) provides participants with information and handouts regarding allergic reactions to laboratory animals, animal bites, and relevant zoonoses based upon the animals used at the worksite during enrollment in the Animal Exposure Program (AEP) and distributes annual reminders.
- The Facility veterinarian presents information on zoonoses and personal hygiene as part of the New Hire/Not Previously Assigned Animal User Orientation process for physical plant staff and to all research personnel identified as animal users.

Other considerations

Primary responsibility for providing training in personal hygiene and proper, ergonomic work techniques specific to particular job assignments and potential for (chemical) contamination rests with the immediate supervisor, animal

facility manager, and the APD as applicable. Animal care contractors are required to provide training to their employees, on a continuing basis, and to provide the project officer with documentation of the training provided.

- Animal care staff receive initial training at hire and refresher training, as needed, on SOPs. Each species-specific (e.g. Rhesus Monkey SOP) and operational (e.g. Dirty Side Cage Wash) SOP describes common hazards in "Section 1. Safety Considerations," PPE requirements, and safe work practices.
- The SciTech Services contract manager and facility veterinarian are NIH designated trainers to administer the "Working Safely with Nonhuman Primates" training course to, respectively, the husbandry and research staff which work with/in NHPs and their areas.
- PIs and or designated senior laboratory staff provide training, supplemented by the ACUC administrator and facility veterinarian, in safe animal handling practices to their research staff.
- The immediate supervisors of animal care staff provide task specific training, e.g. hand restraint of rodents and physical restraint of NHPs including Squirrel monkey pole-tether-collar technique, NHP cage squeeze-back operation, gloved hand NHP capture technique) and certify proficiency of their staff to perform animal procedures.
- The NIDA animal program and Safety Department (coordinated with other DOHS branches) training addresses vivarium related physical hazards including these topics:
  - Compressed Gas Cylinder Safety
  - Ergonomics (Repetitive Motion)
  - Eye/Face Protection
  - Hearing Conservation & Safety
  - Chemical and biological spill clean-up procedures
  - Preventing Hot Weather and Heat Injuries
  - Slips, Trips & Falls – Awareness & Prevention
  - Refer below (Section f), i.) regarding cage-sanitation equipment training.

3) Personal Hygiene [*Guide*, p. 20; *Ag Guide* pp. 4-5]

a) List routine personal protective equipment and work clothing provided and/or required for animal care personnel, research and technical staff, farm employees, etc.

The animal program provides work scrub uniforms for animal care staff and dispenses routine personal protective equipment to all personnel from stations at the entrance(s) of the animal facilities and at or near room entry points for NHP, quarantine, ABSL-2 suite, PET suite, and cage wash.

PPE requirements follow NIH Policy Manual 1340 NIH Occupational Safety and Health Management Program for all laboratory personnel, including those potentially exposed to hazards, 3044-2 Protection of NIH Personnel Who Work with Nonhuman Primates, and the ARAC Guideline: Guidelines for Personnel Protection and Minimum Requirements for Protective Clothing in Animal Facilities. PPE necessary for work assignments is identified in the animal facility's standard operating procedures, in the ASP submitted by the investigator, and if appropriate, by the IBC and DOHS through the NIDA Safety Department before research begins.

NIDA provides these PPE for animal care and animal research staff:

- Scrub work uniform or disposable Spunbound Melt-Blown Synthetic (SMS) laboratory coats to enter occupied animal housing or procedure rooms.
- Steel-toed shoes for husbandry staff.
- Disposable exam gloves (latex, nitrile) to handle animals, soiled cages or equipment, and to enter NHP housing rooms.
- Surgical mask to enter NHP housing rooms or occupied NHP procedure room.
- Bite protective gloves (Kevlar blend, leather) to manually restrain awake NHPs.
- Shoe covers (polyethylene) to enter quarantine and ABSL-2 suites.

Additionally, the following PPE is provided:

- Hearing protection and thermal gloves in the cage wash areas
- Splash protective goggles in cage wash and janitor closets.
- Bouffant caps and surgical masks are provided for use within quarantine and ABSL-2 suites.
- Goggles or face shields are provided if splash risks are present.
- Fit-tested, N95 respirators are provided to participants in the DOHS and Contractor's Respiratory Protection Programs. If medically required or instructed by the NIH IBC for hazard protection, powered air purifying respirators can be provided to animal users.

b) Describe arrangements for laundering work clothing.

Scrub uniforms are laundered by a commercial service; employees may not take laboratory clothing home for laundering. Cloth laboratory coats, used in general laboratories outside of the animal facility, are laundered by an outside, commercial service.

c) Describe provisions and expected practices for washing hands, showering, and changing clothes, including instances where work clothes may be worn

outside the animal facility.

Hands are expected to be washed when exam gloves are changed, removed, or compromised, and on completion of work.

- Hand washing sinks are available in animal testing rooms, change rooms, laboratories, and most of the animal housing rooms.
- Alcohol-based hand sanitizers are available at all exits from animal facilities.

Animal care and research staff are expected to change into dedicated, work scrub uniforms or donning a disposable laboratory jacket upon entry to animal housing and procedures rooms.

- Both Redacted by agreement facilities have men's and women's changing rooms with lockers and showers.
- The animal program provides disposable laboratory jackets to animal users at the entrance(s) of the animal facilities and at or near room entry points to obviate need for clothing change.

Laboratory jackets, scrub uniforms, and other PPE may not be worn outside of the animal facility with these specific exceptions: a clean laboratory coat may be worn over scrub uniforms by the animal care staff and by the veterinary care and breeding staff to travel directly between the Redacted by agreement Redacted by agreement animal facilities.

d) Describe policies regarding eating, drinking, and smoking in animal facilities.

Smoking is not permitted anywhere within any NIH building, except for specifically approved human research studies in rooms with adequate ventilation. Smoking is permitted only in designated areas outside the building.

Eating and drinking are limited to an animal care break room within each animal facility and to personnel offices. No human food or drink is allowed in laboratory or animal areas.

4) Standard Personnel Protection [*Guide*, pp. 21-22]

a) Describe facility design features, equipment and procedures employed to reduce potential for physical injury inherent to animal facilities (e.g., noisy areas, large quantities of chemicals such as disinfectants, ergonomics) or species used (e.g., nonhuman primates, agricultural animals).

Facility design features
NIH requirements for facility design and equipment are specified in the Design Requirements Manual

- The cage wash sanitation rooms have been designed and built to minimize injury potential, such as:
  - Textured methylmethacrylate (MMA) floors reduce slip/fall risks
  - Ceiling mounted exhaust canopies at entrance and exit directs escaping, hot steam away from operators when autoclave and rack washer unload doors are opened.
  - Electric and insulated steam utilities are located within a separate service area between adjacent tunnel and rack washers.
  - Exterior machine controls are contained within a splash proof cabinet.
- Bulk cagewash chemical storage is restricted to dedicated rooms, which are only serviced by commercial vendors, and chemical is pumped through piping in the interstitial space to the facilities' cagewash service closet.

Equipment

- Cage wash equipment supports operator safety.
  Double-door, walk-in type cage washers have:
  - Emergency stop (e-stop) buttons on both the dirty and clean sides of the washers' exteriors and e-stop pull-cables within the interior washing chamber to terminate water flow and wash cycle.
  - Door locks that prevent accidental opening mid-program but also open easily from the inside of the washer to permit emergency egress and wash termination.

  Tunnel washers have:
  - E-stop buttons on both the load and unload end to stop the conveyor belt, as well as,
  - Monitoring sensor at the end of the belt to automatically stop the belt if an item is not removed in time.
- The Redacted by agreement loading dock is equipped with an electric lift within built in ramp to receive ground level deliveries and bridge the gap between truck and platform, and the Triad facility uses a heavy aluminum ramp, as needed, at the building's fixed level dock. Manual and electric pallet jacks are used to move bulk materials, and an electric tug in used in Redacted by for moving animal racks.
- Small quantities of sanitation chemical storage is limited to their point-of-dispensing, typically within janitor closets, commercial chemical storage cabinets, or segregated within general supply storage areas.

Procedures

- PPE reduces potential harm and include:
  - steel toed work shoes,

- hearing protection in the cage wash areas,
- thermal gloves for handling rack washer and autoclaved materials,
- bite resistant gloves for handling and long sleeve laboratory jackets or Tyvek sleeve covers for working with awake nonhuman primates,
- non-latex gloves for those with allergies, and
- splash protective goggles in cagewash and janitor closets.

- Personnel training addresses physical hazard reduction
  - The immediate supervisors of animal care staff provide task specific training, e.g. hand restraint of rodents and physical restraint of NHPs including Squirrel monkey pole-tether-collar technique, NHP cage squeeze-back operation, gloved hand NHP capture technique) and certify proficiency of their staff to perform animal procedures.
  - The NIDA animal program and Safety Department (coordinated with DOHS) training addresses vivarium related physical hazards including these topics:
    - Compressed Gas Cylinder Safety
    - Ergonomics (Repetitive Motion)
    - Eye/Face Protection
    - Hearing Conservation & Safety
    - Preventing Hot Weather and Heat Injuries
    - Slips, Trips & Falls – Awareness & Prevention
    - Refer below (Section f), i.) regarding cage-sanitation equipment training.
- The DOHS has an ergonomic specialist that manages the NIH Ergonomic Program and industrial hygienists to survey occupational noise exposure, heat load levels, and other human factor aspects of cage wash and sanitation operations.

b) Describe likely sources of allergens and facility design features, equipment, and procedures employed to reduce the potential for developing Laboratory Animal Allergies (LAA).

Sources
The likely sources of animal allergens in the NIDA animal care and use program are rats and mice.

Engineering controls
<u>Facility design features</u>
Allergen exposure is reduced with used of 100% non-recycled air and use of negative differential pressures between animal holding area and corridors.

Equipment

The level of allergen exposure is reduced by the use of:

- Microisolator covered cages held within ventilated caging racks whose cage level canopies are exhausted through the rack manifold directly into room exhaust.
- HEPA filtered or bedding change stations,
- Redacted by agreement cagewash vacuum bedding disposal system, and
- HEPA filtered vacuums in animal procedure areas.

Rodent sound attenuating chambers in animal laboratories are exhausted by a small, continuous electric blower and are designed to, when opened, draw air away from personnel at the chamber opening.

Administrative controls

Procedures

Occupational Health and Safety program procedures include:

- Respiratory Protection programs and other allergy related medical services are provided by DOHS and contract staff employers.
    - OMS screening during a pre-placement medical evaluation, counseling and education specifically regarding the potential for animal allergens and preventative measures.
    - Further OHS education and reminders are provided to employees through measures such as: The Office of Animal Care & Use triennial Refresher course for PIs and Animal Users; through posting animal use areas with DOHS 'animal use' signs; and by OMS distributing periodic health and safety information
    - Employees are instructed to report signs and symptoms of animal allergy, to OMS promptly, so that appropriate interventions can be implemented. If the OMS provider suspects that employee may have an allergic reaction to laboratory animal proteins, the complaint is recorded as an occupational illness and
        - Issues the appropriate Workers' Compensation claim forms;
        - Recommends or advises the worker and his/her supervisor that he should temporarily avoid further contact with the animals;
        - Requests that the Safety department specialist evaluate the worksite and review their work practices; and
        - Refers the worker to a community-based allergist for further medical evaluation.
- Animal use areas are posted with warning signs as part of the Laboratory Animal Allergy Prevention Program.

- Species-specific (e.g. Rat SOP) and operational (e.g. Dirty Side Cage Wash SOP) SOPs describe performance of rodent husbandry related procedures which can aerosolize allergens. Husbandry procedures are limited to cage change stations within housing rooms, and PPE is provided for caging sanitation procedures outside of the housing rooms, e.g. cagewash cage/pan dumping/scraping and animal laboratory operant chamber excreta pan cleaning.

PPE

After potential employee exposure to rodent allergen hazards has been reduced to as low as reasonably achievable, the level of exposure is further reduced by appropriately applied PPE which include

- Disposable laboratory jackets for vivarium only use
- Dedicated work uniforms for husbandry staff
- Disposable latex and nitrile gloves
- Surgical or, when medically indicated, N95 particulate respirators are made available.
- Hair bonnets are made available.
- Plastic shoe covers are made available.

c) Describe likely sources of zoonoses and facility design features, equipment, and procedures employed to reduce potential exposure to zoonoses.

Sources

The like source for zoonoses in the NIDA animal care and use program are nonhuman primates.

Engineering controls

Facility design features

NHP housing and testing occurs within rooms along an access controlled corridor. Housing and testing rooms are maintained with negative differential pressure to the corridor.

Equipment

The level of allergen exposure is further reduced by the use of ventilated caging racks, HEPA filtered or bedding change stations, Redacted by agreement cagewash vacuum bedding disposal system, and HEPA filtered vacuums in animal procedure areas.

Administrative controls

Procedures

Purchase specification and quarantine procedures, detailed below in Section III.B.2. Quarantine and Stabilization, screen imported NHPs to exclude carriers of potential zoonoses (e.g. tuberculosis, Herpes B, fecal

pathogens, etc) and the preventative medicine program, described below in Section III.C.1. Surveillance, Diagnosis, Treatment and Control of Disease, continues surveillance of resident NHPs for potential zoonoses.

NHP housing room and corridor sanitation is performed using a tuberculocidal quaternary ammonia chemicals.

Squirrel monkey fMRI and Rhesus MRI staff are enrolled in the AEP as NHP users; procedures to protect others from exposure to potential zoonoses are described in Section I.A.2.b.iii4.e Incidental Animal Contact and Patient Areas.

Animal husbandry and research staff are trained and certified as proficient to perform direct and indirect physical restraint techniques, e.g. hand, pole and collar/tether, restraint chair, squeeze-back operations, and other manipulations to reduce exposure risk from zoonoses.

PPE

After potential employee exposure to zoonotic hazards has been reduced to as low as reasonably achievable, the level of exposure is further reduced by appropriately applied PPE which include:

- Dedicated work uniforms for husbandry staff with availability of Tyvek sleeve covers and disposable laboratory jackets for research staff.
- Surgical masks for entry to NHP housing and testing rooms, a face shield or goggles for entry to Rhesus monkey housing and testing rooms, and, in addition to eye protection, a N95 particulate respirator during housing room based excreta pan scraping and cagewash area pre-sanitation processing.
- Disposable gloves for handling dirty caging and restrained animals and gauntleted, bite resistant leather gloves for handling awake and semi-conscious NHPs.

d) Describe the procedures for the maintenance of protective equipment and how its function is periodically assessed.

The Safety Department coordinates the annual certification of ventilation systems, i.e. biological safety cabinets, cage changing/animal transfer stations, VCR supply air blowers, down draft necropsy tables and chemical fume hoods, surveys of personnel exposure to waste anesthetic gases, and function of clinical x-ray machine and integrity of lead protective garments through contract services and performs regular in-house testing of eye-wash (monthly) stations and deluge shower (quarterly) functions.

The building managers provide annual certification of low oxygen sensors (B1 level MRI areas) and ethylene oxide (B1C923) monitors and monthly monitoring of fire extinguishers.

e) Respiratory Protection

i) Describe situations where respiratory protective equipment is available or required, such as cage washing facilities, feedmills, etc.

Situations involving potential aerosol or airborne hazards are identified by NIDA Safety department reports after routine laboratory surveys or, before research begins, experimental protocol review and by concerned investigators or research support personnel. The NIDA Safety department, aided by OMS and the Respiratory Protection Program manager, as necessary, conducts a comprehensive respiratory hazard assessment to determine the degree of risk, the exposure potential during the specific operation in the work area, and the need for respiratory protection. Additionally, the NIH Institutional Biosafety Committee can identify specific respiratory protective equipment necessary for a particular work assignment written in an ASP.

NIDA SOPs assimilates NIDA guidance documents (ARAC Guidelines for Personnel Protection in Animal Facilities) to require.

- Surgical (dust) masks, which are neither NIOSH certified as a respirator nor require RPP enrollment, are required for entry into housing and testing rooms to protect NHPs. They are also available to supplement the engineering control provided by cage change stations within housing rooms.
- NIOSH approved, N-95 type Aerosol-Removing Respirators are required during aerosol generating activities, e.g. excreta pan scraping and water based, sanitation pre-wash, in Rhesus housing/testing and cage wash areas. Husbandry staff are enrolled in their contractor RPP.

Aerosol-removing, air-purifying, and powered air-purifying type respirators are also available to animal care and use staff where determined by a respiratory hazards assessment.

ii) Describe programs of medical clearance, fit-testing, and training in the proper use and maintenance of respirators.

The Respiratory Protection Program (RPP)is administered by DOHS and an equivalent program provided by contractors' employers. Employees who are required wear a respirator for work at the NIH must be enrolled. NIDA Safety provides appropriate disposable respirators to

personnel entering areas requiring respirators at no cost to the employee. Under this program, enrollees:

- Complete an initial respirator medical clearance questionnaire through the OMS for users of all respirators, including powered air-purifying respirators (PAPRs) and N-95 respirators. The clearance is only required upon initial enrollment.
- Report to the NIDA Safety Department, for quantitative fit-testing after OMS determined the absence of medical concerns that would prevent use of a negative pressure respirator the employee.
- Complete initial, or annual refresher respirator training.
- Schedule and complete initial or annual refresher respirator fit testing.

Employees using dust/mist masks are not required to be included in the RPP. Voluntary use of a NIOSH approved negative pressure, tight fitting respirator (e.g., class N-95 respirator) requires partial participation in the RPP. If DOHS determines that voluntary use will not in itself create a hazard, the supervisor must provide the employee with the written "Information for Employees Using Respirators When Not Required Under the Standard", and the employee must be medically cleared to wear a respirator by OMS.

iii) Describe how such respiratory protective equipment is selected and its function periodically assessed.

Selection
Selection is based on decision tree logic which assesses the immediacy of risk to life or health and the physical state (gas/ particulate) of respiratory hazards and assigns protection factors to different types of respirators. Only NIOSH approved respirators are authorized for mandatory use at the NIH. Several respirators of differing size and type are made available to employees to ensure that wearer acceptability plays a role in selection.

Functional assessment
The NIDA safety specialist or manager fit-tests employees with the same make, model, style, and size respirators to be used during work and repeats fit testing annually or whenever the employee, supervisor, or the OMS reports a change in the employee's physical condition that could affect respirator fit. If the fit of the respirator is reported to be unacceptable, the employee can select a different respirator and be retested.

f) Heavy Equipment and Motorized Vehicles

i) Provide a general list of the types of cage-processing equipment used, such as rack/cage washers, tunnel washers, robotics, and bulk autoclaves. Describe training programs, informational signs, and other program policies designed to ensure personnel safety when working with such equipment.
Note: Details of specific equipment installed in animal facility(ies) are to be provided in **Appendix 15** (Facilities and Equipment for Sanitizing Materials).

Types of cage-processing equipment used

The Redacted by agreement cagewash facilities both contain a tunnel-style cage washer and a pass-thorough rack washer.

Redacted by agreement cagewash facility contains a GarBel waste disposer, and the Redacted by agreement facility contains a vacuum bedding disposal system.

Cagewash areas contain a one bulk autoclave and four pass-thru, standard autoclaves in the Redacted by agreement and one standard load autoclave in the Redacted by agreement The Redacted by agreement standard autoclaves are rarely used; uncommon use of a dirty-side standard autoclave is necessitated by detection of microbial contamination in adjacent quarantine suite or, even rarer, if cages originating from the nearby ASBL-2 suite requires autoclaving for decontamination.

Training programs

Animal care program supervisors train husbandry staff in the safe operation of mechanical cage washers, autoclaves, and sanitizer foamers.

- Only trained animal care staff are authorized to use sanitation/sterilization equipment.
- Manufacturer representatives provide original training on machine operation to the supervisory staff who in turn provides training to animal care staff. Animal care staff training involves:
  - Review of SOPs Clean Side Cagewash Operations and Soiled Side Cagewash Operations which includes practices to manage risks.
  - Demonstration of sanitation/sterilization procedures by trained staff, and follow-on coaching by the supervisor until independent competency is demonstrated.
  - Refresher training is provided and reinforced by periodic rotation of animal care staff to cage wash tasks.

Informational signs

Hearing protection program signs are posted at entry doors to cagewash areas.

Instructional signs are posted on the cagewash equipment to provide emergency instruction and location of the machine's emergency features.

Other cage-processing safety related programs
DOHS and SciTech Services operate other program, as necessary, for individuals:

- Hearing Conservation Program
- Heat Stress Program
- Laboratory Animal Allergy Prevention Program (LAAPP)
- Respiratory Protection Program
- On-site ergonomics evaluation and training

ii) List other heavy equipment such as scrapers, tractors, and farm machinery (manufacturer name, model numbers, etc. are not necessary). Describe training programs, informational signage, and other program policies designed to ensure personnel safety when working with such equipment. *Note:* If preferred, this information may be provided in a Table or additional Appendix.

N/A

iii) If motorized vehicles are used for animal transport, describe how the driver is protected from exposure to hazards such as allergens or zoonoses and decontamination methods employed. Also describe instances where vehicles may be shared between animal and passenger transport.

All intramural animal transport is performed in vehicles which provide a climate-controlled compartment separate from the driver.

- NIDA animal care staff uses the program's vehicle to locally transport rodents that are contained within filtered shipping crates or MI lid covered cages.
- The NIH DVR provides animal transport services between Baltimore and other NIH sites, e.g. the Poolesville, MD NHP quarantine facility, or other Bethesda, MD intramural animal facilities. Dedicated drivers are provided appropriate PPE (e.g. disposable gloves, mask, jump suit, splash shield) for rodents and NHPs.

Long distance arrangements are made with reputable, commercial animal shippers, e.g. World Courier, and rodents are shipped in filtered crates with appropriate rodent chow/gel packs, and bedding.

g) Describe safety procedures for using medical gases and volatile anesthetics, including how waste anesthetic gases are scavenged.

Cylinders of medical gases are required to be secured to an unmovable object, e.g. wall mounted, and capped while stored for use or removal. The NIDA Safety chemical technician is responsible for transporting cylinders, as requested by research personnel, and also assists in regulator selection and placement within a laboratory.

The DOHS TAB administers the NIH Waste Anesthesia Gas (WAG) Surveillance Program. The NIH WAG Surveillance Program identifies and quantifies occupational exposure levels (through surveys and site assessments) every two years to the anesthetic gases used at the NIH and provides information and recommendations for engineering controls and work practices that are effective in minimizing exposures to anesthetic gases.

Waste anesthetic gas is either actively scavenged at the anesthetic machine via connection to the Redacted by agreement building WAG vacuum system or by the use of passive charcoal filter adsorption (e.g. F/Air canisters). Alternatively, volatile anesthetic use is restricted to chemical fume hoods, which are vented directly to the outdoors, or to down-draft tables and other approved local exhaust ventilation (LEV) systems which are vented through the non-recirculating building HVAC exhaust. All LEV devices that are used as the primary means of scavenging are tested and certified by DOHS prior to use with volatile anesthetics.

iii. Animal Experimentation Involving Hazards [*Guide*, pp. 20-21]

1) List, according to each of the categories noted below, hazardous or potentially hazardous agents currently approved to be used in animals that are or will be maintained for more than a few hours following exposure. If the hazardous agent cannot be listed by name for security/proprietary reasons, identify it by the general category of agent and level of hazard.
Note: If preferred, this information may be provided in a Table or additional Appendix.

a) Biological agents, *noting hazard level* (CDC Biohazard Level, Directive 93/88 EEC, CDC or USDA/DHHS Select Agent, etc.). Examples may include bacteria, viruses, viral vectors, parasites, human-origin tissues, etc.

Agents used at **BSL 1**: Adeno-associated virus (AAV)
Agents used at **BSL 2**: Canine adenoviral virus 2 (CAV2), Cholera toxin subunit b (CTb), Lentivirus, Rabies virus (attenuated strain SAD B19 deltaG), rabies viral vector (polymerase double deletion mutant), Picrotoxin
Select Agents (exempt amounts): None

b) Chemical agents, *noting general category* of hazard (toxicant, toxin, irritant, carcinogen, etc.). Examples may include streptozotocin, BrdU, anti-neoplastic drugs, formalin, etc.

MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine), BrdU (5-bromo-2'-deoxyuridine), Daun02, formaldehyde, formalin, 4-hydroxytamoxifen, isoflurane, quinolinic acid, paraformaldehyde, tamoxifen, thapsigargin, urethane.

c) Physical agents (radiation, UV light, magnetic fields, lasers, noise, etc.).

Diode-pumped Solid-State (DPSS) class III or IV lasers (for in vivo stimulation of optogenetic rodent models)
Short life radioisotopes: Carbon-11, Fluorine-18
Long life radioisotopes: Iodine-124, Iodine-125, Carbon-14, Hydrogen-3, and Zinc-65

2) Experiment-Related Hazard Use [*Guide*, pp. 18-19; See also Chapters 2 and 3 in *Occupational Health and Safety in the Care and Use of Research Animals*, NRC 1997].
Note: Written policies and standard operating procedures (SOPs) governing experimentation with hazardous biological, chemical, and physical agents should be available during the site visit.

a) Describe the process used to identify and evaluate experimental hazards. Describe or identify the institutional entity(ies) responsible for ensuring appropriate safety review prior to study initiation.

ACUC ASP Review and Approval process
The Safety Specialist, appointed by the SD as a full voting member of the ACUC, fully participates in ASP review with a primary focus to identify, assess, and mitigate hazards associated with the work described in the ASP. This includes coordinating the review of the ASP and ancillary registration documents or appended safety instructions by the appropriate safety committees (i.e. DRS for the use of radiological agents in animals and IBC for the use of infectious agents and recombinant DNA).

- The PI informs the ACUC of proposed radionuclide, biological agent, hazardous chemical, and recombinant DNA use and lists appropriate biological agent and recombinant DNA registration document numbers in "Section K. Hazardous Agents"; describes safe handling and disposal practices of contaminated animals and associated waste materials in "Section M. Special Concerns or Requirements of the Study"; and identifies locations for performing animal procedures and holding in "Section B. Administrative Data" of the Animal Study Proposal form.

- The animal study may not proceed without Safety Specialist (or Safety Manager in the Specialist's absence) final approval, including confirmation of IBC approval of Registration Documents for the use of recombinant DNA or potential human pathogens.
- Copies of approved ASPs, along with attached (or referenced) registration documents and appended safety instructions, are provided to the designated facility support staff for information and implementation.

NIH Biorisk Review process

ASPs involving infectious disease agents and or recombinant or synthetic nucleic acid research are reviewed concurrently by the ACUC, the NIH Biosafety Officer and the NIH IBC before work may begin.

- The PI electronically registers all work involving human pathogens, non-exempt recombinant or synthetic nucleic acid molecules, human or nonhuman primate blood and body fluids or tissues, or pathogens requiring special permitting with the NIH Biorisk Management Program and the NIH IBC.
- The Biosafety Officer and the IBC conduct an independent biological risk assessment, determine the appropriate combination of practices, facilities and equipment necessary to mitigate the identified hazards and risks.
- IBC decisions and safety requirements are electronically communicated to the PI, and through the Safety Specialist, to appropriate ACUC members.

NIH Radiation Review process

ASPs involving radioactive materials are reviewed concurrently by the ACUC and the NIH DRS before any work may begin.

b) Describe how risks of these hazards are assessed and how procedures are developed to manage the risks. Identify the institutional entity(ies) responsible for reviewing and implementing appropriate safety or containment procedures.

The Safety Department

- Works with Principal Investigators to develop and provide appropriate personnel training, engineering controls, protective equipment, and work practices,
- Verifies that necessary safeguards have been incorporated into study procedures through routine survey of proposed work areas, assurance of appropriate sign postings, participation in semiannual ACUC inspections, and annual survey of registered procedure rooms listed at BSL 1, 2, and 2/3

- Coordinates DOHS TAB annual certification of biosafety cabinets, chemical fume hoods, animal necropsy down-draft tables and other local exhaust equipment is certified annually
- Assesses functionality of eyewashes (monthly) and emergency showers (quarterly)
- Employees or work areas are surveyed for exposure to formaldehyde, anesthetic waste gases, ethylene oxide, noise, heat stress or other potential hazards, as appropriate.

The Division of Radiation Safety (DRS) surveys work sites to monitor occupational exposures and operation of engineering controls.

- Routine monitoring for radioactive contamination of the room and equipment is verified.
- Performs a close-out survey at the end of each study and before the room and equipment are released for use by others.

Redacted by agreement building management monitor, maintain, and repair building ventilation, laboratory waste disposal, electrical systems, and room level oxygen (MRI) and ethylene oxide sensors.

c) Describe the handling, storage, method and frequency of disposal, and final disposal location for hazardous wastes, including infectious, toxic, radioactive carcasses, bedding, cages, medical sharps, and glass.

- Hazardous agent solutions are prepared in general laboratories, and a form of secondary containment is required for transit in elevators or corridors to/from their designated area of use.
- Work surfaces (bench tops, hood surface, etc.) on which hazardous agents/substances are used are covered with plastic trays, dry absorbent plastic backed paper, and/or other impervious material. The protective materials are disposed of after the procedure involving a hazardous substance has been completed.
- Special procedures are performed before (e.g. disinfection, deactivation, etc.) and/or after pick-up (e.g. autoclaving) before they are separately packaged and labeled wastes or caging and enter their appropriate waste stream.
    - Reusable caging is staged and processed *en masse* between the ABSL-2 after decontamination and cage washing.
    - Carcasses are placed in a sealed plastic bag which is placed inside an MPW box for Division of Environmental Protection manager pick up and eventual incineration.

The ORF Division of Environmental Protection provides environmental health and waste management services for general solid, medical pathological, radioactive, chemical, mixed and multi-hazardous wastes

through its Waste and Resource Recovery Branch (WRRB) and oversight through an on-site Environmental Protection manager.

NIH Policy Manual 3032 - Waste Minimization and Management at NIH establishes the policy for management of all types of wastes generated at NIH facilities including and wastewater and references the NIH Waste Disposal Guide as a resource for waste disposal practices.

- The Waste Disposal Guide states that all waste generated in an animal facility with actual or perceived presence of pathogenic agents (e.g., body parts, animal carcasses or tightly packed biological material) must be disposed as medical pathological waste, (MPW). When required, contaminated or biohazardous bedding is placed in Medical Pathological Waste (MPW) boxes, sealed, staged in MPW cold storage room, and screened for radioactive contamination before being released for shipment by the DEP and WRRB personnel. The cleared MPW boxes are then trucked to an EPA-registered, regulated medical waste incinerator.
- The Waste Disposal Guide's "Medical Pathological Waste (MPW) - Management Procedure" section describes the acceptable methods of disposing sharp-related objects. All syringes, needles, scalpel blades, etc., are placed in approved sharps containers, and disposed as MPW.
- Waste fluids are directed to labeled containers for removal by the Environmental Protection manager.
- Procedures for inactivation of infectious agents is general performed via steam autoclave or chemical treatment under the purview the Safety Department.
- Radioactive wastes are also managed in accordance with NRC requirement under the guidance of the ORS Division of Radiation Safety and ORF DEP.

d) Describe aspects of the medical evaluation and preventive health program specifically for personnel potentially exposed to hazardous agents.

NIDA does not conduct work with BSL-3 or BSL-4 agents and only exempted quantities of select agents and toxins (SATs) are used.

Individuals, including contractors, who work in either an ABSL-3 or ABSL-4 facility or with SATs, must enroll in the NIH Biological Surety Program (BSP). BSP participants receive a number of medical services, including AEP enrollment, and are provided informational handouts and a wallet card concerning the pathogens studied in their laboratories. All Individuals enrolled in the BSP are recalled annually. Contract workers, who have contact with animals but are not enrolled in the BSP, receive services equivalent to those included in the AEP from a community-based healthcare provider and their employer coordinates the services.

3) Hazardous Agent Training for Personnel [*Guide*, p. 20]

Describe special qualifications and training of staff involved with the use of hazardous agents in animals.

Special qualifications

- All personnel working with hazardous agents are affiliated with NIDA through a hiring authority or special volunteer agreement.
- NIH Policy (PM 3015, "Admittance of Minors to Hazardous Areas") establishes minimal age qualifications for admittance to research areas and working with hazardous agents.
  - Minors (< age 18 years) may not work with or be present for procedures or research activities involving awake NHPs; may not work with human and/or nonhuman primate blood, body fluids and tissues; or work with radioactive materials. Minors under 18 years of age may not enter a room or area containing a NHP.
  - Minors are excluded from laboratories posted as BSL-2 with BSL-3 practices and from laboratories in which known carcinogens, reproductive toxins or other acutely toxic chemicals are being handled.

Training

NIH has implemented role-based safety and health training requirements. Primary training regarding the handling of hazardous agents is provided by either the DOHS or DRS, to all personnel who might be potentially exposed to hazardous agents in NIH facilities. The NIDA Safety Department coordinates and provides DOHS and DRS training and refresher training on a wide range of OHS subjects. Contract employees commonly attend DOHS training to ensure communication of consistent, institutional standards; however, contract employers provide comparable safety training including OHS training components of an NIH-equivalent, Animal Exposure and Respiratory Protection programs. Required training for all new or otherwise not previously trained laboratory staff, including summer students include:

- Introduction to Laboratory Safety - a web based training course, covers basic laboratory safety in NIH research laboratories and is prerequisite to Laboratory Safety at NIH course. This course introduces laboratory personnel to common hazards and exposure risks including chemical, biological, radiological, and physical hazards that are found in NIH research laboratories.
- Laboratory Safety at the NIH - a three-hour classroom course which presents additional training on the recognition and control of chemical, biological, and physical hazards and provides information on NIH policies and procedures for working safely in research laboratories. Completing the course and its prerequisite meets the OSHA mandated training requirements for a laboratory environment.
- Laboratory Safety Refresher Course - a web-based refresher course providing updates for safety procedures and policies that govern

laboratory safety at the NIH and is required annually of all NIH laboratory personnel.

- Working Safely with HIV and Other Bloodborne Pathogens (for Non-Hospital Personnel) is a two-hour classroom course that describes work practices (including Standard Precautions) in Biosafety Level-2 & -3 laboratories which includes personnel handling non-fixed Old World nonhuman primate tissues. This session meets the OSHA Bloodborne Pathogen Standard training requirement. Bloodborne Pathogen training classes are required only for personnel who work with either primary human blood, body fluids or tissues; primary old world, unfixed NHP blood, body fluids or tissues; or potential pathogens that are infectious to humans via the bloodborne route in the laboratory. BBP training is suggested for ABSL-1 agent use, and required for ABSL- 2 and above agent use.

Primary responsibility for providing training in proper laboratory or animal room techniques specific to a particular facility or job assignment rests with the immediate supervisor, animal facility manager, and the APD as applicable. The DOHS offers formal and informal safety-related training pertaining to animal use as the need arises. Animal care contractors are required to provide training to their employees, on a continuing basis, and to provide the project officer with documentation of the training provided. Specialized OHS training and, for some subjects, annual refresher training is required for staff involved with hazardous agent use including agents used in animals. Research personnel are trained by their PI or lab manager in the specific handling and use of any hazardous agent in animals.

- Radiation Safety (one-on-one interim training is also available)
- Laser Safety Training (Class IIIB & IV lasers)
- Select Agent Program Training (required for handling for non-exempt quantities; only exempt quantities are used at NIDA)
- Working Safely with HIV and Other Bloodborne Pathogens (for Non-Hospital Personnel) on-line course addresses "standard precautions" training for personnel handling only non-fixed old world NHP tissue and are followed under the Pathogen Registration Program.
- In-service, laboratory demonstrations, e.g. Medical Pathological Waste (MPW) disposal practices, proper operation and use of biological safety cabinets, chemical fume hoods, and other primary barrier equipment, as needed.
- The NIH (IBC) can require additional qualifications and training for those who work with biological agents.
- Individuals are required to register with the DRS before beginning work with radioactive materials, and new users must be supervised by an assigned, experience user for storage, use, and disposal of radiologicals. DRS provides training for all personnel (Federal or Contract) working with radioactive materials in animals. Both initial and periodic training in

the safe use of radioactive material is an integral part of the NIH's As Low As is Reasonably Achievable (ALARA) program. Both general and specialized courses are developed to address the nature and types of radioactive material and radiation sources workers may encounter

- Users of select agents, e.g. tetrodotoxin, etc. must register with the NIH Select Agent Program
- The immediate supervisors of animal care staff provide task specific training, e.g. hand restraint of rodents and physical restraint of NHPs including Squirrel monkey pole-tether-collar technique, NHP cage squeeze-back operation, gloved hand NHP capture technique) and certify proficiency of their staff to perform animal procedures.
- In-service presentations are available at the individual lab level covering subjects such as Medical Pathological Waste (MPW) disposal and the proper use of laboratory safety equipment. DOHS representatives are available to collaborate with the Principal Investigator to provide site-specific safety-related training on a case-by-case basis (i.e., high containment, select agents, CPR, AED, shipping biological material, etc.).

In addition, DOHS operates several programs with educational training components which are assimilated into Laboratory Safety at the National Institutes of Health course, and elaborated, as necessary, for individuals:

- Anesthetic Gas Surveillance Program
- Ethylene Oxide Surveillance Program
- Hearing Conservation Program
- Hazard Communication Program (including Chemical Hygiene Plan)
- Heat Stress Program
- Laboratory Animal Allergy Prevention Program (LAAPP)
- Respiratory Protection Program

Optional training offered or available for all staff

- Fire Extinguisher Training
- CPR/AED Training
- Biological Materials Shipper Training
- On-site ergonomics evaluation and training

Further workplace training occurs on a case-by-case basis, as specific-agent use is proposed for either operational or research purposes.

4) Facilities, Equipment and Monitoring [*Guide*, pp. 19-20]

a) Describe locations, rooms, or facilities used to house animals exposed to hazardous agents. Identify each facility according to the hazard(s) and containment levels (if appropriate).
Note: If preferred, information may be provided in a Table or additional

Appendix.

Use of hazardous agents in animals is primarily limited to animal laboratories or designated areas within general laboratories. Two containment suites include areas dedicated to house animals exposed to hazardous agents.

The ABSL-2 Suite is appointed for containment of biological materials, used at the ASBL-1 and ASBL-2 levels, in animal studies.

The ABSL-2 laboratory suite is managed by Redacted by agreement who controls access through entry door proximity card readers, ensures personnel training, monitors suite usage, and coordinates closely with the Safety and animal care program staff.

The ABSL-2 suite consists of Redacted by agreement for transferring animals, staging materials, and donning PPE, Redacted by Redacted by agreement, and a Redacted by agreement Redacted by agreement Redacted by agreement Two cubicles are designated for separate housing of mice or rats; one cubicle contains a certified NuAire Class II, Type A/B3 biological safety cabinet and can also be used for dirty cage staging, and the last cubicle is used for behavioral observation and/or testing and to relocate a function in event of cubicle failure.

Room design/construction features:

All rooms have a minimum ventilation rate of 10 air changes/hour; room pressure is negative to corridors. Exhaust air leaving the ABSL-2 area is filtered through a pre-filter and HEPA filter.

- Floors - Troweled, seamless epoxy resin coved to a height of 6 inches at the wall/floor junction.
- Walls - Epoxy paint finish over filled masonry block and over gypsum panels and metal frame walls.
- Ceilings - Epoxy paint finish over gypsum panels with water resistant fluorescent lighting banks are recessed.

Room air (Redacted by agreement is drawn at the cubicle face, HEPA filtered and supplied, as one-pass air, into the secondary animal enclosure, exhausted from within the cubicle through a pre- and HEPA filter before exiting into the non-recycled building HVAC exhaust system which discharges at roof level, outside of the building and distant from supply air intakes.

The PET laboratory suite is appointed for containment of radiological agents used in animal studies.

The PET laboratory suite is managed by the Biobehavioral Imaging and Molecular Neuropsychopharmacology unit which controls access through entry door proximity card readers, ensures personnel training, monitors suite

usage, and coordinates closely with the Safety, DRS, and the animal care program staff.

The microPET suite consists of 8 rooms: an office [Redacted by agreement]

Redacted by agreement

Room design/construction features:
All rooms have a minimum ventilation rate of 10 air changes/hour; room pressure is negative to corridors.

- Floors - Troweled, seamless epoxy resin coved to a height of 6 inches at the wall/floor junction.
- Walls - Epoxy paint finish over gypsum panels and metal frame walls with exteriorized lead sheet cladding.
- Ceilings – Suspended, scrubable 2'x4' fiberglass reinforced plastic (FRP) tiles with recessed, water resistant LED fluorescent lighting banks.

Suite air exiting into the non-recycled building HVAC exhaust system which discharges at roof level, outside of the building and distant from supply air intakes.

b) Describe circumstances and conditions where animals are housed in rooms outside of dedicated containment facilities (i.e., in standard animal holding rooms). Include practices and procedures used to ensure hazard containment.

Administration of tamoxifen occurs in rodent housing rooms for some experiments. Additionally, rodents are housed in the standard animal holding rooms subsequent to injection of AAV or BrdU injection.

Please refer to Section 4.d, below, for containment practices and procedures used for tamoxifen injected animals.

c) Describe special equipment related to hazard containment; include methods, frequency, and entity(ies) responsible for assessing proper function of such equipment.

At least one item of primary containment is required for all studies involving hazardous agents (e.g. BrdU, adenoviral and lentiviral vectors) within animals. Primary containment may be one or more of the following items:

- Static microisolator cage with Reemay® filter media lid

- Illinois-style Cubicle
- Biological Safety Cabinet or chemical fume hood (for agent preparation and dosing and researcher provided husbandry of cages containing potentially hazardous wastes)

Room ventilation balance, i.e. directionality, is used for secondary containment and is adjusted for air to flow into the animal holding/manipulation area and maintain a negative differential pressure in the room to adjacent corridors and rooms. Types include:

- Standard animal holding and animal procedure rooms have negative differential pressure to corridors.
- The Illinois-style cubicles have negative differential pressure to their room.

General and Animal Laboratories

General and animal laboratories are designed to remain at a negative pressure relative to the corridors. Where applicable, local exhaust ventilation systems, e.g. fume hoods, biological safety cabinets, local exhaust ventilation (LEV), are in place and utilized. Safety eyewashes and deluge showers are located and available for use in general and animal laboratories.

- Acute use of some hazards is routinely performed in animal testing laboratories, e.g. transcranial injection of adeno-associated viral (AAV) vectors though craniotomy.
- Paraformaldehyde and formalin are used in a chemical fume hood or downdraft table Redacted by agreement Redacted by agreement
- Isoflurane anesthetic gases are scavenged through local exhaust ventilation (LEVs) systems to non-recirculating, building HVAC exhaust, waste anesthetic gas vacuum systems, and/or adsorption in canisters (e.g. f/AIR) to minimize personnel exposure. Isoflurane anesthetic gas is also used during tail biopsies, conducted within a cage changing hood and scavenged through adsorption canisters, within the rodent breeding colony housing rooms

Methods and frequency

- Performance of biosafety cabinets, chemical fume hoods, down-draft tables and other local exhaust equipment is certified annually, and functionality of eyewashes (monthly) and emergency showers (quarterly) is regularly assessed. Gas anesthesia vaporizers are certified biannually through a local service company. The Technical Assistance Branch (TAB) monitors waste anesthetic gas exposures during work procedures every two years.
- Employees or work areas are surveyed for exposure to formaldehyde, anesthetic waste gases, ethylene oxide, noise, heat stress or other potential hazards, as appropriate.

- Routine monitoring for radioactive contamination of the room and equipment is verified, and DRS also performs a close-out survey at the end of each study and before the room and equipment are released for use by others.

Responsible entity
The PI is responsible for assessing proper equipment function of vapor chambers.

The Safety Department performs or coordinates DOHS work site surveys to monitor occupational exposures and operation of engineering controls. The Division of Radiation Safety monitors and surveys work site radioactivity use separately.

Redacted by agreement building management monitor, maintain, and repair building ventilation, electrical systems, and room level oxygen (MRI) and ethylene oxide sensors.

d) Describe the husbandry practices in place to ensure personnel safety, including any additional personnel protective equipment used when work assignment involves hazardous agents.

Trained research staff provide husbandry services to animals within containment areas or for the duration of risk from the hazard within standard animal holding rooms.

ABSL-2 Suite

- Rats and mice are housed within the suite for a designated time period after their last injection with a biological agent or hazardous chemical (e.g. lentivirus, adenovirus-recombinant, or canine adenoviral virus, BrdU). Two Illinois-style cubicles are designated for separate housing of mice or rats, and a third cubicle contains a certified NuAire Class II, Type A/B3 biological safety cabinet for servicing opened cages and can also be used to stage dirty caging.
- Once risk from contamination is passed, the research staff transfers the rodents to clean cages and returns the animals to a centralized housing room. Dirty, decontaminated cages are staged within the anteroom; cages are then picked up by Animal Care staff, processed in bulk through the autoclave, if indicated, and the cagewasher before being returned as clean caging to the anteroom.

PET Suite

- Rats and mice (and, in the future,) NHPs are housed for 10 half-lives after their administration with short term (<18.5 hours) radiologic agent. Only "spot changing" of an excessively soiled or wet cage may be performed as sanitation intervals exceed the duration of study.

- Once risk from short-term radiation is passed, the research staff, wearing a face shield, transfers the rodents to clean cages and returns the animals to a standard animal holding room. Dirty cages are scraped and cleaned with Radiacwash before return to the dirty side of cagewash. All dirty bedding, food, and water is entered into the radioactive waste stream. Carcasses, dirty bedding, food, water and disposable cages containing long term (ten half-lives that exceed 4.2 days) radiologic agents enter the radioactive waste stream. Sealed rad-waste boxes are staged within lead-line waste storage receptacles in the PET suite until removed by the DRS contract service.

Standard Animal Holding Room

Trained research staff provide husbandry services to animals within the standard animal holding rooms for the duration of risk from the hazard. Animal are housed in microisolator covered cages and placed onto a VCR or wire-shelf cart. The researcher places hazard warning signs onto animals' cages for animal care to exclude from their routine husbandry practices. Within the room's cage change hood, researcher staff, who supplements standard PPE with a hair bonnet and face mask, transfer animals into a clean cage, transfers bedding waste into a MPW box, and decontaminates caging before returning cage and accessories to dirty side of cage wash for processing. Researchers remove hazard signs from the cage when risk duration expires, and the husbandry staff resumes daily, full animal care.

e) Incidental Animal Contact and Patient Areas

i) List and describe facilities that may be used for both animal- and human-based research or patient areas, including the policies and procedures for human patient protection, facility decontamination, animal transport through common corridors or elevators, and other personnel protection procedures.

A National Institute on Aging (NIA) study involving Macaque species NHPs is conducted in the NIDA [Redacted by agreement] clinical (MRI) research area. Practices are described in the NIA document, "Transportation and Standard Operating Procedures for Magnetic Resonance Imaging of NIA-IRP Old World Non-human Primates in the NIDA Magnetic Imaging Suite Protection & Decontamination," and includes:

- NHPs are screened to verify their pathogen free status (TB, Salmonella/ Campylobacter, Herpes-B), and all personnel, including operators who remain within the MRI control room, are enrolled in an Animal Exposure Program (AEP).
- Single use personnel protective equipment (mucous membrane and hand protection, hair bonnet, and street clothing protective attire) is

used during direct contact with the NHP and soiled materials and when the NHP is present within the MRI scanner room.

- Conscious NHPs are not transported. NHPs are anesthetized within the NIA [Redacted by agreement] vivarium, transported in common corridors through the freight/animal elevator to the NIDA B1 level MRI research area, and returned to the NIA vivarium for anesthetic recovery following the scan. NHPs are transported within an enclosed, stainless steel transport cart with a hinged, locking Lucite side door with small ~ 3" sized mesh covering the air ports; all transporter exterior surfaces are disinfected before exiting vivarium and MRI areas. Signs are posted at the entrances to the MRI suite's corridor to prevent unauthorized entry when NHPs are present.
- Animal procedure supplies, including designated MPW boxes, and dedicated equipment are separate from the segregated human clinical materials.
- Surfaces of the MRI scanning bed are disinfected before and after animal related use, and a barrier of impervious drape material is erected to cover clinical equipment, cabinetry, and floors in the scanning room and in the corridor staging area.
- Disposable materials enter the MPW waste stream at point of use, and equipment is disinfected before removal along with MRI scanner surfaces, countertops and floor at the end of each day's imaging sessions.

Several years ago, NIDA completed studies involving Macaque species nonhuman primates (NHP) in the clinical (MRI) research area. When reinitiated in the future, NIDA will follow similar safeguards as described above.

ii) Describe any *other* circumstances in which animals or caging equipment are transported in common use corridors or elevators (e.g., have the potential to come in contact with individuals not associated with the animal care and use program), and measures taken to mitigate risks associated with such use.

Rodents are transported to general laboratories outside of the vivarium through designated freight elevators. Rodents are contained in cages with secure fitting microisolator lids, and the cages are covered to be obscured from casual view.

Staff is instructed to not wear PPE outside of the animal facility or laboratory during transport to minimize dispersal of allergens to general laboratory corridors.

B. Program Oversight

1. The Role of the IACUC/OB [*Guide*, pp. 24-40]

a. IACUC/OB Composition and Function [*Guide*, pp. 17; 24-25]
Please provide a Committee roster, indicating names, degrees, membership role, and affiliation (e.g., Department/Division) as **Appendix 7**.

i. Describe Committee membership appointment procedures.

Redacted by agreement appoints the ACUC members with delegated authority from the Institutional Official, Dr. Michael Gottesman, NIH Deputy Director for Intramural Research.

ii. Describe frequency of Committee meetings. Note that **Appendix 8** should contain the last two IACUC/OB meeting minutes.

The ACUC meets monthly; additional meetings may be convened to address uncompleted time-sensitive activities or compelling institutional issues.

iii. Describe the orientation, training, and continuing education opportunities for IACUC/OB members. [*Guide*, p. 17]

ACUC members are required to attend the NIH course, "Animal Care and Use Committee Member Training: Defining the Challenge of ACUC Membership." This course is a two-hour interactive lecture course that provides an overview of federal and local policies and regulations that govern animal care and use for the NIH intramural research program, and the ACUC's responsibilities for helping the institute meet these requirements.

Newly appointed ACUC members are required to observe at two to three ACUC meetings before they are permitted to review submissions.

NIDA sponsors ACUC members that attend PRIM&R and AALAS Conferences. Both of our non-affiliated ACUC members and the Regulatory Affairs Officer attended the PRIM&R Conference, March 2017 in New Orleans. Our ACUC Administrator attended the PRIM&R Conference, March 2018 in Columbus, Ohio. NIDA maintains a subscription to NABR. ACUC members receive NABR Updates and Alerts. A monthly list of OLAW, AALAS, USDA-AWIC on-line webinars and seminars is distributed to all ACUC members.

b. Protocol Review [*Guide*, pp. 25-27]
A blank copy of your institution's protocol review form should be provided as **Appendix 9**. Also include forms used for annual renewal, modifications, amendments, etc., as applicable.

i. Describe the process for reviewing and approving animal use. Include descriptions of how:

- the IACUC/OB weighs the potential adverse effects of the study against the potential benefits that may result from the use ("harm-benefit analysis"),
- protocols that have the potential to cause pain or distress to animals are reviewed and alternative methodologies reviewed,
- veterinary input is provided, and
- the use of animals and experimental group sizes are justified.

Note: Make sure you address each of the items above.

Animal Study Proposals (ASPs) and modifications are submitted via email to the ACUC Administrator on a standard form available on the NIDA Animal Program Intranet website.

Full Committee Review Procedures

The ACUC Regulatory Affairs Officer creates an agenda for the convened full committee meeting and assigns primary and secondary reviewers to each submission in consultation with the ACUC Chair, as necessary. Scientific content of all new and renewed protocols involving the use of non-human primates must be approved in advance by the Scientific Director.

Primary and secondary ACUC reviewers are chosen from IRP Sections other than the principal investigator's section. Electronic copies of the ASP are then distributed to the ACUC members along with the agenda, reviewer assignments, and the minutes of the previous meeting seven days prior to the scheduled ACUC meeting.

Primary and secondary reviewers prepare written comments on ASP review forms for presentation to the full committee. The Attending Veterinarian (AV), or his alternate, reviews all ASPs and presents comments to the committee. The Safety representative presents comments to the committee and completes a Safety review form on proposed use of hazardous agents. Input is then solicited from all members of the committee. Following presentation and deliberation of each ASP at the convened full committee meeting, the committee votes on the disposition of the ASP. The committee may vote for one of the following dispositions:

- Approved,
- Tabled for designated member review (DMR) after incorporation of modifications required to secure approval,
- Tabled for full committee review after incorporation of modifications required to secure approval, or
- Disapproved.

An ASP is tabled for designated member review (DMR) only when the ACUC has already obtained sufficient information to judge the humane and appropriate use of animals AND in the absence of a request by any member for full committee review. When a submission is tabled for DMR, the principal investigator (PI) must send an electronic copy of the revised ASP to the Administrator. The revised ASP is distributed to the DMR committee members for review. The reviewers either approve the ASP, require further modifications to secure approval, or request a full committee review.

Following approval by the full committee or DMR committee, the Administrator requests a finalized hardcopy version of the ASP with training assurances and certifications. The signatures of the ACUC chair, the Attending Veterinarian, and others are obtained, and the other review documents and correspondences are collated into a single file folder for each ASP. Written notification of the final approval is sent to the PI, and the Administrator enters the submission into the electronic Animal Care and Use Database to track animal usage. ASP approvals are for three years.

Designated Member Review Procedures

A standing Designated Member Review (DMR) committee is appointed annually at a convened meeting of the ACUC. The current DMR committee is represented by the ACUC Chair, the Attending Veterinarian, the NIDA Regulatory Affairs Officer, and the NIDA Safety Officer.

DMR is employed when the Full Committee votes to table an ASP for DMR. The Chair may add ad hoc members to the committee including those with special expertise. Any ACUC member may request to participate in a DMR committee review.

DMR may also be employed in lieu of a convened meeting. DMR may be granted by the ACUC Chair when there is a justified need for immediate action. This process is infrequently used. A copy of the ASP (or modification) is sent to all members of the committee. The members are requested to respond with either approval for designated member review or to request full committee review. Members are given 2 business days to respond. If any member requests full committee review then the ASP must be reviewed at a convened meeting of the ACUC. If no members request full committee review the DMR may proceed with the review of the ASP (or ASP modification). The designated member reviewers either approve the ASP, require modifications to secure approval, or request a full committee review.

Harm-benefit Analysis

- The committee members understand the need to weigh the objectives of the study against potential animal welfare concerns and have been trained in the concept of the three R's.
- The ACUC encourages open discussion at all convened meetings during review of protocols. The ACUC has engaged in many discussions and formed subcommittees to address various subjects including food and fluid restriction, physical restraint, behavioral testing, and pain relief.
- Members of the NIDA-ACUC frequently serve on NIH Policy and Guideline committees addressing these issues.
- The ACUC has experienced veterinarians present at all meetings to provide their input, the scientists on the ACUC are very experienced in contemporary behavioral methodologies, and the non-affiliated members contribute frequently to the discussions and serve as reviewers.

Review of potential to cause pain or distress

The NIDA-ACUC has established "Guidelines for Assigning Pain and Distress Categories in Research Animals" which it uses in review of procedures involving potential for pain and distress.
For all USDA category D and category E procedures (rodents included), the ACUC requires the PI to certify in the ASP (ASP, Section N, item 5) that no valid alternatives were identified based on a review of pertinent scientific literature. Scientific justification is required to explain why the use of anesthetics, analgesics, sedatives or tranquilizers during or following a painful procedure is contraindicated for column E procedures (ASP, Section H and column E justification).

The Attending Veterinarian (AV) is involved in reviewing all potentially painful procedures as part of the ACUC's review of ASPs. The committee seeks the advice of the AV whenever there is a question of potential pain. The veterinarians will pre-review ASPs and provide guidance to PIs on anesthetics/analgesics and methods to reduce or alleviate pain. The Facility Veterinarian is involved with daily care of all animal cases, prescribes treatment including pain relief, and notifies the AV if there are concerns with pain and distress.

The PI must describe the methods and sources used in the search; database references must include the databases searched, the date of the search, period covered, and keywords used and description of keywords/search strategy using the applicable Boolean operators. In consultation with the NIH research librarian, the ACUC provides written search instructions, including prescribing a minimum number of keywords with applicable Boolean operator terms, and recommends pertinent NLM databases to support the PI to describe the methods and sources used to review the literature including personal communications with subject-expert consultants.

Animal Selection and Experimental Group Size

ASP Section E requires a rationale for animal use, justification of the appropriateness of each species selected, and a justification of the number of animals to be used and basis for group size (s) determination. Group sizes are generally based on previous experience and the type of statistical analysis to be used.

ii. Describe the process for reviewing and approving amendments, modifications, and revised protocols. If applicable, include a description/definition of "major" vs. "minor" amendments.
Note: If preferred, this information may be provided in a Table or additional Appendix.

The ACUC "Policy for Review of Significant Changes to Animal Study Proposals" conforms with NIH ARAC "Guideline Regarding Significant Changes to Animal Study Proposals" and current OLAW guidance (NOT-OD-14-126).

All proposed changes are recorded by cumulative revision and review of the previously approved Animal Study Proposal. The Significant Changes Policy describes the changes that are considered significant and minor. Significant changes are organized into subcategories which determine the method of review and approval. The following provides a general summary of the ACUC policy content.

- The ACUC mandates Full Committee or Designated Member Review Committee for significant changes which include proposing changes in Principal Investigator, species, procedures or animal strain with potential to result in greater pain, distress, invasiveness, and other adverse outcome, and addition of a novel study area location.
- The Attending Veterinarian or Facility Veterinarian may approve conditional administrative changes which include proposing alternate or additional numbers of animals and or strain, clinical or experimental drug regimens, euthanasia method, food/water regulation regimens, and changes to surgical procedures which are consistent with applicable policies and guidelines and not expected to impact substantially and directly on the health and well-being of the experimental animals or introduce risks to personnel.
- Minor changes, which include addition of personnel, change of study rooms within areas already under ACUC oversight, and editorial corrections, may be approved by the ACUC administrator.

The ACUC is informed of all changes to ASPs which were approved subsequent to its last meeting. Documentation of the review and approval is documented in the ACUC minutes and filed with the ASP.

c. Special Considerations for IACUC/OB Review [*Guide*, pp. 5; 27-33]

i. Experimental and Humane Endpoints [*Guide*, pp. 27-28]

1) Describe the IACUC/OB's review of "humane endpoints," i.e., alternatives to experimental endpoints to prevent or in response to unrelieved animal pain and distress.

Endpoint Determination
NIDA follows the NIH ARAC Guideline, Guidelines for Endpoints in Animal Study Proposals which promotes refinement to minimize pain or distress. NIDA also uses an internal ACUC Guideline, Guidance on Selecting Doses of Investigational Drugs, for establishing dose ranges to anticipate outcomes from treatment with novel drugs, to develop alternate humane endpoints, and to refine dosages for the study. The veterinarian and the ACUC assist investigator in determining appropriate humane endpoints, as needed, at the time of protocol development and review.

The Principal Investigator specifies alternative experimental endpoint criteria in ASP Section F for preemptive euthanasia or removal from study to prevent, terminate, or relieve pain or distress when procedures are potentially lethal and or expected to cause significant symptomatology. The PI indicates the expected effects and time course, the observation frequency, identifies the personnel responsible for evaluation, and provides criteria for preemptive euthanasia. (e.g. transcardial perfusion, or removal from the study).

2) For studies in which humane alternative endpoints are not available, describe the IACUC/OB's consideration of animal monitoring and other means used to minimize pain and distress (e.g., pilot studies, special monitoring, other alternatives).

When endpoints are not available
- The ACUC may approve a pilot study in a subset of animals to define morbidity, time course of effects, and appropriate observation frequency to develop effective endpoints prior to approval of an entire study.
- The ACUC may require a veterinarian be present to observe the initial procedure(s) to confirm adequacy of proposed endpoints, refine the proposed endpoints, or suggest other ameliorative interventions.

3) Identify personnel responsible for monitoring animals for potential pain and distress and describe any mechanisms in place to ensure that the personnel have received appropriate species- and study-specific training.

The investigative, veterinary, and animal care staff perform both general and study-specific observations of research animals.

The animal care staff are trained to recognize signs of illness and pain by the facility veterinarian and contractor training program. Research staff are trained though the NIH course "Using Animals in NIH Intramural Research: Guidelines for Animal Users" regarding species-specific and by their PI regarding study-specific recognition of pain and distress.

ii. Unexpected Outcomes that Affect Animal Well-being [*Guide*, pp. 28-29]
Describe how unexpected outcomes of experimental procedures (e.g., unexpected morbidity or mortality, unanticipated phenotypes in genetically-modified animals) are identified, interpreted, and reported to the IACUC/OB.

As part of the daily standard animal health assessment, the veterinary care staff identifies deviations from anticipated experimental and spontaneous outcomes described in the ASP. Within the ASP, the PI indicates whether genetically-modified animals have any known associated deleterious phenotypes, and if so, indicates alternative endpoints for these animals.

When any unexpected morbidity, moribundity, or mortality is reported or identified by the facility veterinarian, the facility veterinarian evaluates the situation, contacts the investigator, informs the APD who in turn notifies the ACUC Chair. The investigator applies his/her scientific judgment to determine whether unexpected outcomes could be related to the study and proposes solutions in collaboration with the facility veterinarian. Significant outcomes might require amendment of the ASP and ACUC approval before continuing the study. The PI may amend the study to avoid the outcome, propose a standard treatment plan, or special husbandry practices to prevent further reoccurrence. If efficient intervention would alter experimental results, then a scientific justification may be required and animals may be placed in USDA category E.

All rodent breeding at NIDA is centralized and performed by breeder technician specialists with training from Jackson Laboratories. Within the breeding colony unanticipated phenotypes in the GM rodents are identified by the breeding specialists and reported to the Technical Director of the Transgenic Facility. The Technical Director investigates any new line imported or created (through direct genetic engineering or through crossing) for:

- Any already known phenotype that might affect animal wellbeing.
- Any suspected phenotype based on literature or experience on similar lines
- Any other phenotype especially if it might affect animal wellbeing.

- New lines are especially scrutinized and monitored. In some cases, the breeding colony staff can conduct specific monitoring such as weight monitoring. Most genetically modified mouse lines have been imported and already have a known history of any existing phenotype abnormalities.

- Most genetically modified rat lines have been designed and created for NIDA investigators and are therefore more closely monitored over several generations to detect potential unexpected outcomes.
- The Technical Director monitors the reproductive performance and health of rodent breeders and their offspring to identify phenotypes that negatively impact well-being.
- The Technical Director reports unexpected outcomes and adverse phenotypes to the ACUC through the Breeding ASP modification and the annual review process. Reports include any required management of adverse phenotypes by
  - Special husbandry procedures such as powdered/dough diet, provision of supplemental gel water source, and floor-level condiment cup feeders
  - Altered breeding schemes such as breeding phenotypically normal heterozygotes, and
  - Accelerated culling (euthanasia) of offspring which are not expected to meet experimental use specification, e.g. sex, cohort size, genotype before expression of adverse outcomes.
  - Special monitoring.
- A database is maintained on all abnormal phenotypes in the colony.

iii. Physical Restraint [*Guide*, pp. 29-30]

Note: This section is to include only those protocols that require prolonged restraint. Brief restraint for the purpose of performing routine clinical or experimental procedures need not be described.

1) Briefly describe the policies for the use of physical restraint procedures or devices. Include, if applicable, the IACUC/OB definition of "prolonged."

The ACUC's Animal Restraint Policy:

1. The Animal Study Proposal must include (1) the justification for the restraint, (2) a description of the restraint device, (3) the minimum duration of restraint required to accomplish the research objectives, (4) the training/habituation procedures, (5) the observation intervals, and (6) criteria for removal from restraint..
2. The least restrictive means of restraint consistent with the aims of the study should be used. Use of prolonged restraint, including chairing of nonhuman primates, should be avoided unless it is essential for achieving research objectives and is specifically approved by the ACUC. Systems that do not limit the animal's ability to make postural adjustments should be used when compatible with protocol objectives.
3. Animals that fail to adapt must be removed from the study. The disposition of these animals should also be indicated in the ASP alternative endpoints.
4. Restraint devices are not to be considered normal methods of housing, and must be justified in the animal use protocol.

5. Restraint devices should not be used simply as a convenience in handling or managing animals.
6. The period of restraint should be the minimum required to accomplish research objectives.
7. Alternatives to physical restraint should be considered and documented in the literature search where appropriate for animals in USDA category D or E.
8. Animals to be placed in restraint devices should be allowed to habituate or given training with positive reinforcement to adapt to the equipment and personnel.
9. Devices should be suitable in size, design, and operation to minimize discomfort, pain, distress, and potential for injury to the animal or the research staff. For example, the size and weight of head mounts, especially for mice, that are used to connect to tubing during testing should be evaluated as part of a restraint system and should be made as small and light as possible, as not to interfere (burden) with normal head postural adjustments.
10. Veterinary care should be provided if lesions or illnesses associated with restraint are observed. The presence of lesions, illness, or severe behavioral change often necessitates temporary or permanent removal of the animal from restraint.
11. The purpose of the restraint and its duration, and recognition of pain/distress signs associated with restraint should be clearly explained to all personnel involved with the study.

Prolonged restraint is defined as greater than 1 hour if not acclimated, and greater than 4 hours if acclimated

2) Describe animal restraint devices that are used or have been used within the last three years. For each device, briefly describe

- the duration of confinement
- acclimation procedures
- monitoring procedures
- criteria for removing animals that do not adapt or acclimate, and
- provision of veterinary care for animals with adverse clinical consequences.

Note: If preferred, this information may be provided in a Table or additional Appendix.

Method of Restraint	**Species**	**Approved Duration of Restraint**
Chair/Pole & collar	Squirrel monkey	Up to 2 hours
Chair/Pole & collar/Head post	Rhesus monkey	Up to 4 hours
Plastic body tube	Rat	5-20 minutes

fMRI: Head mount	Rat	Up to 2 hours
Pupillometry: Head restraint	Mouse	Up to 1 hour
Restraint Stress: Mouse restrainer tube	Mouse	4 hours

Chair restraint

Acclimation: Squirrel monkeys are fitted with collar and chain and Rhesus monkeys are fitted with collars. After habituation to wearing their collar (with chain for Squirrel monkeys) in their home cage for several days, training for pole-chain-collar or pole-collar restraint is done over a 3-5-day period for Squirrel monkeys and 2-4 week period for Rhesus monkeys. All monkeys are further habituated to the restraint sequence and the transport box prior to the chair training. The monkey is placed into the chair for short periods of time once daily. The duration of chair restraint is increased over several days depending on the monkey's adaptation. Positive reinforcement with pellets are provided while in the transfer box and chair.

Monitoring: Monkeys are continuously monitored during the behavioral sessions via individual video camera monitoring for well-being and any maladaptive behavior during the entire testing sessions. Computer monitoring equipment shows real time behavioral responses during the testing sessions and time out periods. Continued appropriate behavioral responses indicate the monkey is not having any difficulty. The operator is always present in the testing room or immediate area and can hear the audible clicks and visually inspect if there are any problems. The restraint chairs have been used extensively for over 30 years by the NIDA investigators without untoward effects. Direct contact with and close observation of the subject occurs immediately before and after the experimental session.

Removal criteria: Training will be temporarily suspended if untoward behavioral reactions occur. Failure to progress in habituation after a full week of training will require longer periods of time on a particular step or backtracking to the previous step.

Veterinary care is provided if lesions or problems associated with restraint are observed, and the animal is temporarily or permanently removed from restraint procedures. Skin lesions are permitted to heal fully before re-application of collar or placement in the chair.

Plastic body tube

Acclimation: Rats are placed in a clear, vented plastic tube fitted with a tail slot to prevent unnatural body postures. This procedure is needed to produce reinstatement of drug seeking and has the advantage, over alternative procedures, because it is simple and fast, and only causes moderate, temporary discomfort to the animal. The tube, designed to restrict movement, is placed on an absorbent pad to alleviate moisture buildup.

Monitoring: Animals are monitored throughout the procedure to ensure that no physical harm results from movement and temperature is monitored to prevent hyperthermia.
Removal criteria: In the event that respiratory distress (highly unlikely because the tubes have several holes/gaps for ventilation), sustained struggling effort or unnatural body postures occur, the subject is immediately removed from the restrainer

fMRI
Acclimation: Rats are habituated to handling, hand restraint, and being placed in a jacket. The period of restraint is gradually increased to 2 hours. For acclimation to head restraint the rats are lightly anesthetized with isoflurane for immobilization while head mount is secured. Duration of restraint starts with 1-2 minutes and is extended to 2 hours with daily training over 6-week period.
Monitoring: Temperature, respiratory rate, and heart rate are monitored as sensitive indicators of stress which have been correlated with blood corticosterone levels in other studies.
Removal criteria: Animals that fail to acclimate are removed from the study. If a head mount becomes loose the animal is euthanized.
The veterinarian is contacted if there are any health issues related to the procedures.

Pupillometry:
Acclimation: Mice are gradually acclimated to head-fixed restraint for habituation to the setup over at least 3 days or until the mouse tolerates the head restraint (maximum of 6 days). If the mouse tolerates head restraint well, the time interval of immobilization will be gradually increased over 3 days: 5 minutes on first day, 10 minutes on second day, and 15 minutes on third day. To calm the mouse and create a positive association with the head restraint process, the mouse will receive a few drops of sweetened water (0.1% saccharin or 10% sucrose). The relevant behavioral experiment will start only after the mouse is acclimated. Head restraint duration will be the minimum needed to accomplish the scientific objective (assay basal, auditory cue-driven, and drug-induced pupil dilation).
Monitoring: After mice are placed in head restraint, they will be monitored continually. If the mouse shows signs of distress (including struggling, vocalizations, increased respiration rate, or escape behavior) lasting 2 minutes, the mouse will receive another drop of sweetened water. If the distress persists for another 2 minutes, the acclimation session will end and the mouse will be returned to home cage. This will be repeated on the following days.
Removal criteria: If a mouse fails to tolerate the head restraint after 6 days of acclimation trials, it will be taken out of the experiment and euthanized.

Restraint Stress:

Acclimation: Acclimation is not possible as this procedure is used as a stressor to study the effects of stress on the development of psychiatric disorders such as schizophrenia and depression. Mice are placed in plastic tubes which restrict their movement for four hours a day, for 14 consecutive days.
Monitoring: During the initial trials, mouse body temperature is carefully monitored to ensure that mice won't become hyperthermic. Mice are monitored daily for signs of illness (lack of grooming, loss of body weight, etc.) and if any show signs, the veterinarian is consulted. Mice are monitored daily for signs of morbidity: rapid breathing; shallow, labored breathing; weight loss (more than 15% of body weight loss); ruffled fur (rough hair coat); hunched posture; hypothermia or hyperthermia; ulceration dermatitis or infected tumors; diarrhea; impaired ambulation; evidence of muscle atrophy or other signs of emaciation; bleeding; inability to remain upright.
Removal criteria: If signs of morbidity are observed over the course of the experiments, the veterinarian will be notified and, if necessary, mice will be excluded from the study and euthanized using CO2 asphyxiation followed by cervical dislocation.

iv. Multiple Survival Surgical Procedures [*Guide*, p. 30]
Note: One survival surgical procedure followed by a non-survival procedure is not included in this category.

1) Describe the IACUC/OB's expectations regarding multiple survival surgery (major or minor) on a single animal.

NIDA Policy is that multiple survival surgical procedures on a single animal must be scientifically justified and approved by the ACUC.

The ACUC apply these criteria for approval of proposed multiple survival surgery:
- Multiple, survival surgeries must be scientifically related and required components of a research proposal.
- Multiple, major survival surgeries may be done if needed for clinical reasons.
- Animal well-being must be considered and outcomes evaluated.
- Cost savings is not an adequate reason for performing multiple procedures

2) Summarize the types of protocols currently approved that involve multiple major survival surgical procedures

Note: If preferred, this information may be provided in a Table or additional Appendix.

Sequential craniotomies: There are many rat and mouse studies in which intracranial injection of viral vectors or tracer molecules is followed by

intracranial implantation of cannulas for insertion of optogenetic fibers, microscopic imaging cameras (e.g. two photon microscopy), or microelectrodes for behaving rodents. These studies require craniotomies at two time points to allow incubation of the virus or the movement of tracers. The time between surgeries varies with the study and may be 2 days to 4 weeks. The investigator is responsible for postoperative monitoring following each surgical intervention on each animal and documents this on post-operative care card for three days.

The ACUC also approves multiple minor surgeries to remove non-functional intravenous jugular vein catheter and re-implant the same or opposite side jugular vein in rats. In squirrel monkeys the internal jugular, femoral, and external iliac veins may be re-implanted. Re-implantation of catheters is approved in order to ensure the completion of ongoing behavioral studies.

v. Food and Fluid Regulation [*Guide*, pp. 30-31]. *Note:* This does not include pre-surgical fast.

Summarize the types of protocols that require food and/or fluid regulation or restriction, including:

- justification
- species involved
- length and type of food/fluid regulation
- animal health monitoring procedures and frequency (e.g., body weight, blood urea nitrogen, urine/fecal output, food/fluid consumption)
- methods of ensuring adequate nutrition and hydration during the regulated period

Note: If preferred, this information may be provided in a Table or additional Appendix.

In many of the behavioral studies at NIDA, either food or water, restriction or regulation, is used to motivate animals during training to press levers to obtain reward pellets and facilitate self-administration of drugs of abuse to ensure consistent behavioral performance during experimental sessions.

In some rat studies, food restriction is used for weight maintenance where obesity can interfere with study measurements.

Rats and mice – Food restricted

- The most common procedure used in rodents is food restriction which involves the provision of a standard quantity ration once a day with the total amount adjusted to maintain target body weight at not less than 85% of pre-restriction, ad libitum, body weight.

- Body weights are recorded a minimum of once weekly and more frequently in the initial restriction phase until target weight is reached and stabilized.
- Special feeding log is initialed daily.
- Daily ration must provide at least 30% of ad libitum calories but is typically 50-70% of ad libitum calories per week.
- Male rats must be 325 gm body weight prior to initiation of food restriction and females 275 gm.

Rats and mice– Water regulated access

- Scheduled access to water is provided once daily for periods ranging from 10 to 120 minutes.
- Rodents are introduced to the new schedule gradually over a period of several days.
- Body weights are recorded daily.
- Skin turgor and appearance are evaluated daily.
- A maximum of 10% body weight loss is permitted, otherwise access to water access is reinstated.

Squirrel monkeys- Food restricted

- Food is restricted to maintain a target body weight which is limited to 85% of ad libitum weight.
- Food and water consumption amounts are recorded daily. Biscuit quantities are adjusted accordingly.
- Body weights are recorded daily, Monday through Friday.
- Physical examination with hematocrit and serum chemistry is performed quarterly.
- Daily ration must provide at least 30% of ad libitum calories but is typically 50-70% of ad libitum calories per week.
- A body weight loss of 25gm below the target weight which is unresponsive to increased feed requires notification of the veterinarian
- When clinically indicated, ad libitum feeding is reestablished until the animal exceeds target weight and or reaches pre-restriction weight.

Rhesus monkeys- Water restricted (approved, not yet initiated)

- Rhesus monkeys are given the largest amount of fluid possible while still achieving research goals. The amount of fluid restriction is customized to the individual animal.
- Animals is fluid-restricted 6 days a week for the duration of cognitive training and testing. A vacation day is permitted on Saturdays.
- Prior to the implementation of fluid restriction, each animal undergoes a health assessment by Animal Program (AP) veterinary staff.
- Baseline weight is established. The baseline weight is reassessed quarterly in growing animals.
- Physical examination with hematocrit and serum chemistry is performed quarterly.

- Changes in the amount of administered fluids are done gradually over the course of a week at a time, plus or minus 20 ml/kg/day, to avoid physiological stress caused by acute dehydration.
- The initial daily target fluid-regulation is set to approximately 30 ml/kg/day of the baseline body weight. Animals do not go below 20 ml/kg/day of fluid.
- Each animal is observed daily during fluid regulation. Food intake, consistency of stool, skin turgor, behavior of the animal, and a qualitative assessment of urine output are recorded daily.
- Mature animals on fluid regulation are weighed at least once a week. If animals that are still growing, they will be weighed three times a week. No animals younger than four years of age are used on fluid restriction.
- Weight loss of more than 10% of baseline weight is reported to the veterinary staff.
- Signs of dehydration such as decreased food intake, little or no urine production, scant hard dry feces, lethargy, sunken eyes, increased skin turgor or behavioral changes is reported to the AP veterinary staff.

For all animals

- Operant testing procedures can be sensitive indicators of physiologic and behavioral health, and disruption of consistent responding may prompt temporary or permanent removal of animals from study.
- Animal Care staff reviews investigator room logs to confirm daily provision of food and water and reports deviations daily including suspicion of inappetence from leftover ration for food restricted animals.

vi. Use of Non-Pharmaceutical-Grade Drugs and Other Substances [*Guide*, p. 31]
Describe the IACUC/OB's expectations regarding the justification for using non-pharmaceutical-grade drugs or other substances, if applicable.

The ACUC reviews scientists' justification for use non-pharmaceutical-grade chemicals within the Animal Study Proposal (ASP), Section F. Description of Experimental Design and Animal Procedures.

- The ARAC Guidelines for the Use of Non-Pharmaceutical Compounds is used for the development of ASPs involving non-pharmaceutical-grade substances.
- The NIDA species-specific SOP Section 7, Drug Administration provides guidance regarding grade/purity of substances, formulation of the final product, and issues such as sterility, pyrogenicity, stability, pH, osmolality, site/route of administration, physiological compatibility, and quality control.

Non-pharmaceutical-grade substances are used only when FDA approved, USP/NF, or BP pharmaceutical grade compounds are not available, and, analytical grade chemicals, as scientifically appropriate, are used. A licensed compounding pharmacist from the central NIDA pharmacy prepares controlled substances, including anesthetics and experimental drugs, for dispensing, and the central pharmacy provides a compounding service for scientists who prefer to not

compound their own non-controlled formulations. Some scientists also prepare solutions of experimental drugs with clean materials and sterile vehicle in sterile containers in laboratory drug preparation areas.

vii. Field Investigations [*Guide*, p. 32]

Describe any additional considerations used by the IACUC/OB when reviewing field investigations of animals (non-domesticated vertebrate species), if applicable.

Not applicable.

viii. Animal Reuse [*Guide*, p. 5]

1) Describe institutional policies regarding, and oversight of, animal reuse (i.e., on multiple teaching or research protocols).

Principal Investigators do not reuse animals as a strategy to reduce the total number of animals necessary for a given study. The overwhelming majority of animals are acquired or transferred without a history of prior research use.

A small number of rodents, which may have been prepared and or tested by extramural collaborators, can be imported and used on NIDA ASPs to support collaborative, primarily imaging-based, research. Additionally, nonhuman primates, which may have been used by other intramural institutes, are occasionally acquired when history of prior research use is compatible with the NIDA scientific objectives, as determined by the principal investigator and the Scientific Director. The ACUC reviews the justification for use of animals that have already undergone experimental procedures; the ASP form- Section G Survival Surgery also explicitly queries whether survival surgery was performed on any animal prior to being placed on a study.

Internally, the Squirrel monkey animal model involves parenteral and intravenous administration, via chronically implanted intravenous catheters, of drugs and lever pressing within an operant chamber. Squirrel monkeys are commonly enrolled in sequentially approved renewals of animal study proposals over the five to twenty-plus years of residency within the colony. Suitability to enroll and continue within a study is determined the veterinarian, in consultation with scientists, by health assessments prior to starting, at quarterly intervals during, and after completion of active (i.e. daily testing) use.

When rodent species are transferred between protocols to initiate studies on experimentally naïve or collaboratively extend studies on experimentally prepared rodents, the investigator submits an Animal Transfer form, which references originating and recipient ASP numbers and a brief description of prior and planned manipulations, to the Animal Care supervisor. The transfer form is forwarded to the ACUC administrator to confirm that the species, strain, or model is approved in the receiving ASP and that there are sufficient

numbers of animals left on the protocol to deduct the amount transferred. If reuse is a concern, the veterinarian will evaluate further and may refer the request to the ACUC Chair for guidance.

2) Briefly describe the types of activities currently approved that involve the reuse of individual animals.
Note: A list of specific protocols involving reuse of animals should be available during the site visit.

Animal Study Proposals
The same Squirrel monkeys may be transferred between ASPs and subsequent renewals, as their studies require the same highly trained Squirrel monkey animal model for similar, specific scientific objectives.

Naïve rats and mice are frequently transferred between investigators. Many of the research ASPs use same transgenic lines generated in the centralized breeding colony.

Breeder colony excess animals may be used for hands-on training new research staff in animal handling and basic biomethodology classes. Under this protocol, individual animals are used for demonstrating IP and SQ injections no more than three times in one week.

3) Describe other instances where the final disposition of animals following study does not involve euthanasia, including adoption, re-homing, rehabilitation, etc.
Note: A list of specific protocols involving reuse of animals should be available during the site visit.

Nearly all transfers of rodents are to collaborators at extramural institutions involving donation of rats and mice bred under auspices of the centralized breeding colony's ASP after completion of appropriate Material Transfer Agreements.

A memo from the NIH Deputy Director for Intramural Research provides policy guidance on the "adoption" of animals from NIH. To date, two rats have been transferred through a written Animal Adoption Agreement to an animal research employee as pets.

NIDA does not have any specific protocols designating reuse of animals.

2. Post-Approval Monitoring [*Guide*, pp. 33-34]

a. Describe mechanisms for IACUC/OB review of ongoing studies and periodic proposal/protocol reviews (e.g., annual, biennial, triennial, or other frequency).

The ACUC has implemented a post-approval monitoring program. The program is referred to as the NIDA Protocol Adherence Verification (PAV) program. In this program, currently active protocol(s) from a selected laboratory are evaluated against actual animal user awareness and procedural adherence. The procedure involves three components: (1) a meeting with all animal users listed on the protocol(s), (2) an observation of procedures by one or two animal users performing surgery, behavioral tests, tissue collection, etc. and (3) a visit to the animal housing area for animals under the protocol(s). Prior to the meeting, a copy of the protocol(s) and the PAV SOP, including all questions to be asked, are emailed to the PI. A subset of the ACUC (usually 3 members, including a scientific member on a rotating basis) attend all parts of the lab PAV visit. At the procedure observation, the ACUC members observe the surgery or other procedure approved on the protocol. Finally, a visit to the lab's animal housing areas is done. This program captures information that verifies that procedures being performed are consistent with those approved in the protocol, that individuals who work with animals are appropriately trained and qualified to perform technical, hands-on procedures, that animal body weight and postoperative records are being kept. In addition, the inspection of anesthetic, analgesic, euthanasia and test drug expiration date status, evaluation of PPE and surgical instrument sterilization practices, and inspection of housed animals' general health, environmental enrichment, and social housing status are checked. Deviations are corrected on the spot. If modifications to protocols are required, the PI is notified at the time of the visits. The ACUC Chair reviews the PAV visit results and proposes corrective actions. The PI receives a letter stating the results, and the results are presented at the monthly ACUC meetings. The frequency of PAV visits has been two per month, with a recent change to one a month approved by NIH OACU. The PAV team asks the lab members if they have any questions or concerns regarding ACUC or animal program processes.

The ACUC approves Animal Study Proposals (ASPs) for 3 years but requires submission of an Annual Review form for Full Committee review. The PI is provided with the number of animals used to date, and the Annual Review form also prompts PIs to report intent to continue activities; to assess changes and to submit, as appropriate, ASP modifications; to provide a description of any adverse or unanticipated events; and to update the emergency contact list.

The ACUC can approve resubmitted ASPs for a three-year renewal after full committee review; however resubmitted ASPs are subjected to current ACUC requirements and applicable regulations, policies, and guidelines that have been implemented or revised since the prior approval, during their review process.

Adverse and unanticipated events are reported to the ACUC through a chain of communication beginning with the care staff. All animals are observed twice daily by trained animal care staff, all health issues are reported to the veterinarian and added to

the electronic distribution of morbidity and mortality reports, the veterinarian follows up on evaluation of outcomes of animal procedures with investigators, the APD is informed, and the ACUC Chair. Supervisors report all employee work related incidents involving occupational health and safety to the Safety office for follow up.

b. Describe the process and frequency with which the IACUC/OB reviews the program of animal care and use.

The Program Review is conducted semi-annually by a subcommittee of the ACUC. All members of the ACUC are invited to participate. Review and discussion is open for all aspects of the Program. Agenda items are requested in advance. The committee uses the Guide for the Care and Use of Laboratory Animals (8th edition), the Animal Welfare Act Regulations, the PHS Policy on Humane Care and Use of Laboratory Animals, and the AVMA Guidelines on Euthanasia of Animals (2013) as the basis for the review. The subcommittee reports the results of the review at a convened meeting of the ACUC.

c. Describe the process and frequency with which the IACUC/OB conducts facility and laboratory inspections.

- Describe the rationale or criteria used for exempting or varying the frequency of reviewing satellite holding facilities and/or animal use areas.
- If contract facilities or contractor-provided personnel are used, describe procedures used by the IACUC/OB to review such programs and facilities.

Note: A copy of the last report of these reviews should be included as Appendix 10.

The ACUC performs semiannual review and inspection of the animal facilities and review of the animal care and use program in the Spring and Fall each year. Facility inspections include all rooms in the animal facilities and all animal use areas inside and outside the animal facilities including acute procedure areas and satellite areas. Members of the committee are assembled in teams to inspect assigned areas. The teams consist of at least two ACUC members but typically teams have 3-4 members each. The ACUC inspects laboratory room sanitation logs, post-operative care records, surgical practices, fluids and drugs used in animals, and anesthetic and scavenging apparatus, as part of their semi-annual inspection. During ACUC inspections, animal behavioral testing chambers are randomly swabbed to monitor effectiveness of research staff sanitation procedures. Comments from all members are collected and collated into the final report. Any deficiencies found are written and sent to the responsible party with request for correction and date for correction. Formal responses are kept on file in the ACUC Administrator's office and corrections are verified by the Animal Program Director.

NIDA does not use any contract facilities

d. If applicable, summarize deficiencies noted during external regulatory inspections within the past three years (e.g., funding agencies, government, or other regulatory agencies)

and describe institutional responses to those deficiencies. *Note:* Copies of all such inspection reports (if available) should be available for review by the site visitors.

As a Federal research facility, the USDA does not inspect the animal program. USDA reporting requirements are centralized through NIH.

e. Describe any other monitoring mechanisms or procedures used to facilitate ongoing protocol assessment and compliance, if applicable.

Controlled Substances
All DEA Schedule I-V drugs are ordered, received, recorded on inventory, and stored by the centralized NIDA IRP pharmacy. The pharmacy dispenses drugs and records the transfer to an authorized recipient. The recipient maintains an inventory which includes: 1) Name of controlled substance, 2) Unit size received, 3) Amount received, 4) Date used, 5) Signature of user, 6) amount on hand. The NIDA pharmacy performs a physical inventory, confirms storage locations, and reviews scientists' dispensing logs at least twice a year.

3. Investigating and Reporting Animal Welfare Concerns [*Guide*, pp. 23-24]
Describe institutional methods for reporting and investigating animal welfare concerns.

Individuals are encouraged to report animal welfare concerns to any of the following individuals: the IO, Director OACU, the ACUC Chair, members of the ACUC, or the veterinarians. Reporting can be done anonymously, and government whistleblower policies are observed. All personnel are informed of the methods to report during orientation training and the IO's memo "Communicating Animal Care and Use Concerns within the NIH Intramural Research Program" which is posted at the entrances to both animal facilities and on the Animal Program's internal webpage.

- All animal welfare concerns are directed to the ACUC Chair.
- The Chair initiates appropriate steps to validate the real or potential seriousness of the issue/allegation and documents for the record the time and date of notification, who notified the Chair, and who else was notified.
- The Office of Animal Care and Use (OACU) is notified as soon as the Chair has determined a reportable incident has occurred. OACU may render an opinion upon receiving the Chair's initial report whether the issue/allegation appears to be an incident that should be reported to the Office of Laboratory Animal Welfare (OLAW).
- If appropriate, the Chair appoints an investigative agent or subcommittee from the ACUC membership to investigate the issue/allegation.
- The investigative agent or subcommittee conducts investigation, formulates recommendations, and reports back to the ACUC.
- At a convened meeting with a quorum present, the ACUC then deliberates the investigative report & associated recommendations, formulates any additional conditions and/or corrective actions, and either:
  a) awaits further information from the subject of the investigation;
  b) awaits further information from the investigative body; or

c) closes out the investigation and renders a final report.

- Final reports (destined for reporting to OLAW) are delivered to the Office of Animal Care and Use (OACU) for final routing and disposition to the IO.
- Institutional actions to remedy reportable incidents or noncompliance are reported back to the reporting individual by the ACUC Chair.

Concerns originating from outside the NIH are handled by an NIH ombudsman. A policy established by the NIH in 1992 appoints a Senior NIH scientist to function as the ombudsman

4. Disaster Planning and Emergency Preparedness [*Guide* p. 35]

Briefly describe the plan for responding to a disaster potentially impacting the animal care and use program:

- Identify those institutional components and personnel which would participate in the response.
- Briefly describe provisions for addressing animal needs and minimizing impact to animal welfare.

Note: A copy of disaster plan(s) impacting the animal care and use program must be available for review by the site visitors.

The NIDA Animal Program Disaster Response Plan:

- Identifies scenarios, to which personnel and animals may be vulnerable to harmful consequences, and more routine events that would trigger need for immediate responses to emergencies impacting animals. Includes centrally managed housing areas, animal laboratories within the centrally managed vivaria which house rodents for more than 24 hours, and a single laboratory outside of the vivarium. Scenarios include, but not limited to, events such as electrical outages, HVAC system component failures, fires, mechanical equipment breakdowns, building and local water delivery system failures (including scheduled shut down of HVAC and water for physical plant maintenance and repair) and flooding (including hurricane inundation), Baltimore City's highest ranked natural hazard.
- Tabulates scenarios with outlined measures to prepare for, respond to, and recover from emergencies including provisions for:
  - Room level, climate control equipment, i.e. portable, electric air chillers and oil-filled heaters with power extension cords and water hoses, are stored and staff is trained in their deployment and use.
  - Supplies for initial shelter-in-place (SIP) practices, i.e. 110 ready-to-eat meals, 15 sleeping blankets, and inflatable mattresses, are stored for SIP personnel use and replenished.
  - Light Sources, e.g. flashlights, headlamps, are routinely distributed to all animal holding areas, and backup batteries and charged two-way radios are maintained charged for emergency communication within the vivaria.
  - The animal program, air-conditioned, animal transport van, routinely used for transporting rodents and supplies between NIDA buildings, is maintained in

readiness for use; additional NIDA trucks are available for transporting supplies between NIDA buildings and vendors.
    - Euthanasia supplies, i.e. additional CO2 cylinders for rodent euthanasia and sufficient volume of injectable drugs for NHPs, are stocked according to required time and material estimates for typical animal population size if mass euthanasia is necessary.
    - Capture nets, injection pole, and bite resistant gauntleted gloves are present for escaped NHPs.
    - Personal Protective Equipment, e.g. Tyvek jumpsuit, surgical and N95 masks, disposable gloves, disposable laboratory jackets, shoe covers, face shields and goggles, and additional work scrub uniforms are stocked for increased use during emergencies and to anticipate delay in replenishment.
    - Routine storage of up to two weeks of food and capacity to store additional quantities to anticipate delivery interruption. The DVR animal program has a larger, on campus, food storage capacity, and has limited ability to supplement other IRP programs in emergency or disaster response circumstances.
    - Quantities of potable water required daily is estimated for typical animal population size, and the Redacted by agreement reverse osmosis water system can provide potable water at a reduced capacity but can be prioritized to animal use.
- Identifies the Animal Program Director as the authority for making decisions impacting animal care and use, designating key personnel to execute response tasks and organizing response and recovery actions including elective euthanasia of animals. Key animal program personnel are members of the NIDA IRP SendWordNow, a web-based alert notification system, and of the NIH-Animal Contact lists to receive immediate notification of emergency events through AlertNIH notification system.
- Describes general plans addressing:
    - Minimal standards of animal husbandry and veterinary care during abnormal operating conditions to ensure the health and welfare of animals.
    - Procedures for mass euthanasia of animals.
    - Animal evacuation resources including contact points for the NIH Animal Response Team and nearby, similar entities with potential to rapidly enter, and ad hoc aid and support agreement.
    - Prioritization of animals for preservation through shelter-in-place, relocation, evacuation, or euthanasia.
- References additional emergency event guidance resources, developed by NIH for intramural institutes, for responding to additional unexpected emergency/disaster scenarios.

Participating Institutional components and personnel

NIDA's animal program needs and planned responses is locally implemented but is integrated into the NIDA disaster plan and aligns with the trans-NIH Disaster Plan, also known as the Continuity of Operations Plan (COOP), developed by the Disaster Response Animal Advisory Committee (DRAAC). Personnel participate in disaster responses at the intramural research program, NIDA, and NIH levels.

IRP level

- The IRP Crisis Response Team is composed of leadership for separate Emergency Support Teams for animal resources, information technology, research activities, and administrative support composed of security, environmental health and safety, logistics (transportation), facilities, and contracting/procurement personnel.
    - All government and, through their statement of work, contract animal care staff are designated as Tier I, Emergency Employees, and are required to work during federal government closures (e.g. lapse of funding) or other unexpected conditions (e.g. inclement weather) or events. The animal care contractor, SciTech Services, can provide skilled and experienced animal care staff from other contract, when feasible, to muster adequate numbers for an acute surge in labor requirements.
    - The IRP Crisis Response Team leader serves as liaison with the NIDA Emergency Coordinator and NIH Animal Response Team.
- Support assets continue to participate.
    - Building security contract staff are present on site all hours of the day and seven days a week.
    - NIH Occupational Medical Services' physicians remain on call to facilitate and support care provided by Bayview Medical Center emergency room and on-campus, urgent care health providers.
    - Baltimore City Fire/EMS is located adjacent to the Bayview campus.
    - Signs are mounted in centralized managed facility and the Redacted by agreement satellite animal facility to display names and telephone numbers of the primary and backup veterinarians, facility supervisors, IRP Security, and building maintenance engineers in case of after-hours requirements for veterinary or other emergency services.

NIDA Level

- The NIDA Emergency Coordinator is responsible for coordinating all emergency response and recovery activities within NIDA and is the main point of contact for the COOP/Disaster Recovery Coordinator when the COOP is activated.
    - The IC Emergency Coordinator functions as a conduit between the NIDA Leadership Team, the NIDA Crisis Response Team (CRT), and the NIH Disaster Recovery Coordinator.

NIH Level

- The NIH COOP is designed to ensure continuity of the NIH mission essential functions. The NIH Director is responsible for determining whether an event requires a COOP activation. The NIH Disaster Recovery Coordinator directs the Continuity of Operations Program, managed by the NIH Division of Emergency Coordination and Preparedness, and remains the focal communication point for the nine COOP Emergency Support Teams (EST) which respond to an event and coordinate NIH resources. ESTs include Safety, public information, logistics, information technology, administrative support, facilities, and the animal resources team.
    - The Animal Resources Team (ART) is comprised of key members of the NIH Office of Animal Care and Use (OACU) staff and experienced NIH animal program managers and veterinarians. The on-call ART coordinator is accessed through email, text, and phone.

- The ART would be activated to assist the IRP CRT and manage contingent NIH resources only when a catastrophic event occurs that exhausts the resources/abilities for the IRP animal program to continue to operate.
- The ART coordinator is the point of contact if communication with external entities, e.g. USDA, FEMA, MEMA (Maryland), etc. is necessary.
    - The ART includes the NIDA IRP animal program in its website based roster of points-of-contact information and other critical information such as species, number of rooms, facility hazards, security systems, etc. which remains accessible to COOP EST and conducts semiannual tests of the NIH Alert system for those in the ACU programs.

II. Animal Environment, Housing and Management

Note: Complete each section including, where applicable, procedures performed in farm settings, field studies, aquatic environments, etc.

A. Animal Environment

Note: Facility-specific details regarding mechanical system construction and operation is requested in Section IV.B.5. and **Appendix 11**; current (measured ***within the last 12 months***), detailed (by room) performance data must also be provided as indicated in **Appendix 11.**

1. Temperature and Humidity [*Guide*, pp. 43-45]

a. Describe the methods and frequencies of assessing, monitoring, and documenting that animal room or housing area temperature and humidity is appropriate for each species. *Note:* If preferred, this information may be provided in a Table or additional Appendix.

Assessing

Temperature and humidity of animal housing and procedure rooms within the central vivarium and a designated satellite housing area are continuously assessed through a Building Automation Systems (BAS).

Any person can to report abnormal environmental conditions in animal housing and laboratories by contacting either the veterinarian, animal facility manager, or the building engineer. Emergency contact phone numbers are posted at the entrances to both facilities.

Monitoring

The BAS notifies Redacted by agreement building engineers and animal care supervisors through electronic messaging of excursions exceeding alarm points:

	Building Engineers	Animal Care Supervisors
Rats and Mice	< 69°F and > 75°F	< 64°F and > 76°F
Squirrel Monkey	< 75°F and > 81°F	< 72°F and > 82°F
Rhesus Monkey	< 73°F and > 79°F	< 70°F and > 80°F

Automated notification is also sent to building engineers if humidity exceeds 70% R.H.

Documenting
The BAS electronically records temperature and humidity at frequent intervals and archives several weeks of data.

Each building's animal care staff generates and reviews a daily report containing the prior 24-hour high, low, average temperatures, and humidity.

- Deviations are identified in the "Facility Maintenance" section of the "Daily A.M. Report" report and distributed through electronic mail to animal program management.
- Work orders are initiated, as appropriate, to investigate and correct deviations.

Current, detailed (by room) performance data is provided in Appendix 11. Heating, Ventilation, and Air Conditioning (HVAC) System Summary.

b. List, by species, set-points and daily fluctuations considered acceptable for animal holding room temperature and relative humidity.
Note: If preferred, this information may be provided in a Table or additional Appendix. [*Guide*, pp. 44 and 139-140]

Temperature and humidity is controlled at the room level within a +/-2° F range of species specific set points and within a target range of 35-55% relative humidity.

	Temperature Set-point	Humidity Set-point
Rats and Mice	72° F	45%
Squirrel monkey	78° F	45%
Rhesus monkey	76° F	45%

c. Temperature set-points in animal housing rooms and/or environmental conditions are often outside of the species-specific thermoneutral zone. Describe the process for enabling behavioral thermoregulation (e.g., nesting material, shelter, etc.) or other means used to ensure that animals can control their thermoregulatory environment. Include a description of IACUC/OB approved exceptions, if applicable. [*Guide*, p. 43]

Resources and processes are provided to allow animals to control their temperature and avoid cold or heat stress:

- Daily temperature fluctuations are kept to a minimum and temperature settings in housing and testing areas are aligned to avoid repeated large demands on the animals' metabolic and behavioral processes to compensate for changes in the thermal environment.
- Rodents are socially housed, whenever possible or unless exempted by ASP, to support behavioral adaptation mechanisms.
- Rodents are provided slotted cage floors to prevent animal wetting and evaporative chilling when housed within operant testing chambers.
- Rodents in solid bottom cages are provided a deep layer of absorbent and insulating, fine hardwood contact bedding.

- Mice are provided insulating nesting material unless exempted by ASP.
- Rats are provided paper tunnel shelters, unless exempted by ASP, and breeding rats are also provided crinkled paper nesting material.
- Primary enclosures are constructed from or contain materials with low heat conduction properties, e.g. polycarbonate rodent cages.

ACUC Exemption
There are no ACUC approved exemptions to temperature set-points in animal holding rooms.

PIs request exemption from provision of nesting material and shelters within the cage in Section M of the ASP form. The ACUC extensively reviewed potential impact from structural cage enrichment on behavioral neuroscience animal studies: *Altered Animal Housing and Standardization of Results: A Review of Literature Concerning Changes in Animal Housing and Its Consequences – A Report to the NIDA ACUC* (Redacted by agreement Redacted by agreement

Exemptions to social housing is described below, II.B.2.b.ii.

2. Ventilation and Air Quality [*Guide*, pp. 45-47]

a. Describe the methods and frequencies of assessing, monitoring, and documenting the animal room ventilation rates and pressure gradients (with respect to adjacent areas). *Note:* If preferred, this information may be provided in a Table or additional Appendix.

The HVAC system provides continuous air volumes at 10 air exchanges per hour or greater in all animal housing rooms. Housing, testing, and procedure rooms for animals are maintained under negative pressure gradients and personnel areas are maintained under positive pressure gradient to animal service corridors.

Assessing
- Automated building systems provide continuous monitoring of supply and exhaust air flows, and, in the Redacted by agreement supply and exhaust boxes are yoked to continually maintain room pressure gradients.
- Air flow measurements are performed triennially by a commercial air balancing vendor to verify the automated building systems' performance.

Monitoring
- Real time performance data (e.g. supply and exhaust CFM) is accessible from a BAS terminal to the animal care staff and monitored by building engineers. Automated notification is also sent to Redacted by agreement engineers if a room's air flow drops by 25% from its given setting.
- Animal care staff monitors air pressure gradient semiannually at rooms' doors using a smoke pen and as needed.

Documenting
- The BAS electronically displays air flow data and records automated notification events.

- Recorded smoke testing results are recorded and used to generate an electronic work order to correct deviations from the specifications and to confirm corrections.
- The commercial air balancing vendor documents findings and any subsequent adjustments.

Current, detailed (by room) performance data is provided in Appendix 11. Heating, Ventilation, and Air Conditioning (HVAC) System Summary.

b. Describe ventilation aspects of any special primary enclosures using forced ventilation.

Primary enclosures are placed within two types of secondary enclosures which use forced ventilation: sound attenuating chambers containing an operant testing chamber and alcohol/drug vapor inhalation racks containing a standard polycarbonate cage.

- Squirrel and Rhesus monkeys are held in behavioral and operant testing devices within sound attenuating chambers during daily experimental sessions; some rodents can be held within sound attenuating chambers for several weeks within vivarium testing rooms. Sound attenuating chambers typically contain multiple, baffled openings within the chamber walls to allow passive infiltration of room air as enclosure air is exhausted by a small, continuous electric blower. Chambers are designed to provide approximately 10 air exchanges per hour and, when opened, to draw air away from personnel at the chamber opening.
- An alcohol/drug vapor inhalation rack (La Jolla Alcohol Research, Inc.) is a large, clear plastic cabinet with passive exhaust from the rear of the cabinet to draw air away from personnel when the hinged front doors and individual inhalation chambers are opened. The rack contains six clear plastic inhalation chambers (25.75" x 19.25" x 12.25" interior) with a hinged, gasketed front panel and a separate port for forced room air (15 LPM resulting in approximately nine air exchanges per hour). Supply air passes through the heated alcohol vaporizer to enter the chamber and a separate port for air to passively exit the chamber directly to the room HVAC exhaust and without any recirculation. The inhalation rack is equipped with vacuum pump and chemical scrubber for to actively remove aerosolized drugs from exhausted air. Rodents are housed within standard, static polycarbonate cages with a perforated lid (microisolator lids whose Reemay® filter media was removed) for the duration of the study which may be several months. The electric air pump is designed for several years of continuous operation, is powered by [Redacted by agreement] emergency electric supply. A loud battery powered alarm located in the corridor is activated if the pressure of the forced supply air falls to less than half of the normal setting.

A description of the dimensions and composition of the primary chambers are described in the Primary Enclosures and Animal Space Provisions appendix.

c. If any supply air used in a room or primary enclosure is recycled, describe the percent and source of the air and how gaseous and particulate contaminants are removed.

Supply air to housing and testing rooms is 100% fresh, 95% filtered air, and is not recycled.

Ventilated racks supply individual rodent cages with HEPA filtered air which originate from a mixture of fresh room air and highly diluted cage air which escaped from capture into the building's exhaust system.

Sound attenuating chambers are also passively ventilated with room air containing air exhausted from other chambers whose gases and particulates are diluted to insignificant levels by large volume of fresh room air.

3. Life Support Systems for Aquatic Species [*Guide*, pp. 84-87]

a. Provide a general description of institutional requirements for enclosures using water as the primary environmental medium for a species (e.g., aquatics).

Aquatic species are not housed.

b. Provide a general description of overall system(s) design, housing densities, and water treatment, maintenance, and quality assurance that are used to ensure species appropriateness.
Note: Facility-specific tank design and parameter monitoring frequencies should be summarized in **Appendix 12** (Aquatic Systems Summary).

Aquatic species are not housed.

4. Noise and Vibration [*Guide*, pp. 49-50]
Describe facility design features and other methods used to control, reduce, or prevent excessive noise and vibration in the animal facility.

Facility Design
Exterior noise abatement
- Outdoor sources are muted due to below grade location of Redacted by agreement animal facilities.

Interior noise abatement
Airborne transmission
- Spaces for noise-generating mechanical HVAC systems, boilers, generators, bulk trash compactors are located on separate floors.
- Spaces for noise-generating fMRI/MRI, elevators, cage wash operations, and nonhuman primates are isolated by corridors and with heavy-grade, gypsum drywalls with sound-attenuating battens or concrete block walls, which extend to the concrete slab above.

- Husbandry and corridor operations noise sources are compartmentalized by:
  - Automatically closing doors at cagewash and breeding colony corridor entries.
  - Sound-attenuating, corridor doors with mechanical closers, door frames with rubber bumpers and elastomeric gaskets, and automatic-drop, door sill closures.
  - Double-door, entry vestibule for the Redacted by agreement breeding area.
  - Ventilated cage rack mounted blowers contain rubber isolation pads. The number of operating motors has been reduced in Redacted by agreement mouse and Redacted by agreemen rat breeding rooms by replacing individual rack mounted blowers with a single, floor located air blower (MultiPlex™, LabProducts) with a manifold to distribute air to racks.
  - Polyurethane or cushioned rubber casters are used on rolling stock, and rack casters are lubricated after cage wash sanitation to reduce corridor circulation noise.
  - Rodents and NHPs are placed within sound attenuating chambers for behavioral testing and background, "white noise" may be supplied within operant chambers. Open speaker radios are not permitted in housing rooms.
  - Fire alarm annunciators and public address speakers are located within corridors, outside of housing and testing rooms to minimize potential animal disturbance; fire alarm drills are scheduled and coordinated through the animal program to avoid disruption of behavioral studies.

Impact transmission

- Heavy-grade drywall or suspended ceiling panels cover an interstitial space containing wiring, utility distribution pipes, and HVAC ductwork to mitigate noise transmitted from the concrete slab of the floor immediately above the vivarium.
- Noise originating from building repair and maintenance work, e.g. drilling, demolition, renovating, is scheduled, coordinated through the animal program, and communicated to scientists to avoid disruption of behavioral studies.

B. Animal Housing (all terrestrial, flighted, and aquatic species)

1. Primary Enclosures

Note: A description of primary enclosures used (e.g., cages (conventional, individually-ventilated cage systems (IVCS), etc.), pens, stalls, pastures, aviaries, tanks) should be included in **Appendix 13**.

a. Describe considerations, performance criteria and guiding documents (e.g. *Guide*, *Ag Guide*, ETS 123 and/or other applicable standards) used by the IACUC/OB to verify adequacy of space provided for all research animals, including traditional laboratory animal species, agricultural animals, aquatic species, and wildlife when reviewing biomedical, field and agricultural research studies.

The *Guide* and Animal Welfare Regulations (AWR) are used as the basis for selection of cage size and housing density.

- Guidance in cage type selection and housing density is provided in each species-specific SOP Section 3, Animal Receipt.
- Overcrowded cages are identified and remediated by the animal care staff following SOP Section 10, Cage and Rack Changing.
- For breeding rodents, one female is removed from a trio before the first litter falls; cage size is adequate to contain up to two adults with a single litter.
- Squirrel monkeys are housed in group 2 or greater sized cages which exceed individual weight based space requirements, and Rhesus monkeys are housed in group 4 sized cages.

b. Describe space exceptions to the guiding documents (*Guide*, *Ag Guide*, ETS 123, and/or applicable standards), indicating the references, considerations and performance criteria used (e.g., by the IACUC/OB) to verify adequacy of space provided for all animal species covered by the program. [*Guide*, pp. 55-63]

There are no current space exceptions for housing.

If deviation from the Guide or AWRs is needed, the ACUC would review the need for and type of adjustment proposed in an Animal Study Proposal or NIDA SOP and base their determination on performance outcomes with due consideration to the Guide/AWRs, professional judgment, survey of literature and current practices, scientific requirements of the study, risks to animal welfare, and the animal's physical and behavioral needs.

2. Environmental Enrichment, Social, and Behavioral Management [*Guide*, pp. 52-55; 63-65: *Ag Guide*, Chapter 4]

a. Environmental Enrichment

i. Describe the structural elements of the environment of primary enclosures that may enhance the well-being of animals housed (e.g., resting boards, privacy areas, shelves/perches, swings, hammocks).

The following structural elements are used in primary enclosures:
Rodents
- Solid bottom caging is standard housing for rodents,
- Plastic dome shelter (Bioserve) for breeding mice,
- Tunnel (paper tube, Landy Resources, LLC, Potomac, MD) for rats,
- Levers, which are integral to operant chambers used, for long term, rodent housing in dedicated testing rooms within the vivarium.

Nonhuman primates
- Elevated perch rod.
- Opaque partitions as partial visual barriers between neighboring animals.

ii. Describe nonstructural provisions to encourage animals to exhibit species typical activity patterns (e.g., exercise, gnawing, access to pens, opportunity for exploration, control over environment, foraging, denning, burrowing, nesting materials, toys/manipulanda, browsing, grazing, rooting, climbing).

These nonstructural elements are provided to encourage exhibition of species typical activities:

Rodents

- Suspended feeders with food biscuits and, for rats, paper tunnel to stimulate gnawing behaviors,
- Contact, hardwood chip bedding, and nesting materials, accommodate natural burrowing behavior,
- Nesting materials, i.e. compressed cotton squares (Nestlet, Ancare Corp, Bellmore, NY) for mice and crinkle paper (Enviro-dri®, Shepherd Specialty Papers, Watertown, TN) for breeding rats to promote nesting behaviors and to control their temperature,
- Group housing of rodents is promoted whenever not precluded by protocol to permit affiliative social and thermoregulating huddling behaviors.

Nonhuman primates

- Manipulata to encourage play behaviors,
- Elevated feeders and perches to encourage vertical activity.
- Group housing of animals is promoted whenever not precluded by protocol to permit affiliative social behaviors.
- Cage racks are positioned to facilitate visual and auditory communication.

Many on-study animals receive pellets and other rewards during their operant testing activities which also provide enriching manipulative activity and cognitive challenge

b. Social Environment [*Guide*, p. 64]

i. Describe institutional expectations or strategies for social housing of animals.

NIDA follows the NIH ARAC Guidelines for Social Housing of Rodents and Aquatic Species. In brief, NHP's and rodents are socially housed unless precluded by ACUC approved ASP (in its Section M, Special Concerns or Requirements of the Study), ACUC approved species-specific SOP, or veterinary care considerations.

Cages of varied floor space are provided for rat housing, and social housing of growing rats can be maintained by transfer to a larger cage when compatible with study requirements.

Evaluation of suitability of non-exempted, individual NHPs for social housing is made by a senior primate caretaker, and animals are tested and acclimated for

group housing as neighboring animals for several days before attempting social housing. Initially continuous observation (up to 2 hours) and then intermittent observation (every 10 min, up to 2 hour) observation of social behavior is made followed by daytime trials during provision of standard diet and high value, food based enrichment and, later, overnight trials to verify the group's compatibility.

ii. Describe exceptions to these expectations (e.g., veterinary care, social incompatibility) and other typical justification approved by the IACUC/OB for housing animals individually.

Typical exceptions to social housing include demonstrated social incompatibility, and veterinary health and husbandry considerations such as illness, aggression, or breeding requirements.

The ACUC summarizes major justifications in their document "Justification for Individual Housing Non-human Primates in Long-term Behavioral Studies" and in species specific SOP Section 15: Social Housing and Environmental Enrichment Plan. PIs request and justify exemption from social housing based on study design or cagemate attrition within the cage in Section M of the ASP form.

Typical justification approved by the ACUC for housing animals individually include:

- Scientific requirements for stable, behavioral testing performances, not only from one session to the next, but also over long periods in excess of one year, preclude social housing.
- Scientific requirements to measure and control quantities and timing of food access and, in rodents, also water access, for each individual animal preclude social housing.
- Risks from damage to externalized, chronic intravascular catheters, and, in rodents, also optrodes, optical probes, cannulae, electrodes, etc for animal health and disruption of experimental research may preclude social housing

iii. Describe steps taken with isolated or individually housed animals to compensate for the absence of other animals (interaction with humans, environmental enrichment, etc.).

- In the Animal Study Proposal form, Section M, Special Concerns or Requirements of the Study, instructs PIs to elect inclusion/exclusion other elements of the species' environmental enrichment plan.
- Daily interaction with research technicians (e.g. gentling) and animal care staff (e.g. routine husbandry) and pellets and other rewards during their behavioral testing sessions provide additional compensation for reduced social enrichment from a conspecific.

c. Enrichment, Social and Behavioral Management Program Review [*Guide*, pp. 58, 69]

Describe how enrichment programs and exceptions to social housing of social species are regularly reviewed to ensure that they are beneficial to animal well-being and consistent with the goals of animal use.

- The ACUC evaluates the justification for exceptions to social housing for consistency with the scientific goals of animal use during review of initial submission and modifications of the Animal Study Proposal.
- The ACUC evaluates the execution of enrichment program and exceptions to social housing during semiannual animal facility inspection.

d. Procedural Habituation and Training of Animals [*Guide*, pp. 64-65]

Describe how animals are habituated to routine husbandry or experimental procedures, when possible, to assist animals to better cope with their environment by reducing stress associated with novel procedures or people.

Habituation to handling, injection, and connecting cannulas and catheters to infusion/ sampling tubing is a common practice of researchers to reduce unwanted stress in animals involved with behavioral testing, and its positive influence carries over to routine husbandry procedures.

Animal care and research staff are consistently assigned to the same rodent and NHP housing rooms to facilitate staff to recognize species and individual animal's daily behaviors and to acclimate NHPs to interaction with individual staff members.

NIDA ACUC policies, Standard Operating Procedures, and Animal Study Proposals describe training practices to acclimate animals to restraint devices and personnel. Rhesus monkeys are acclimated to wear a plastic or metal collar, and squirrel monkeys are progressively acclimated to wear a plastic or leather collar, fabric jacket, and a fine, tethered chain. They are progressively trained to the pole-collar (Rhesus) or pole-tether-collar (Squirrel monkey) technique for brief restraint during transfer between cage, transport box (Squirrel monkey), and chair, between cages for husbandry, and, with addition of positive reinforcement, to chair restraint for operant testing. Positive reinforcement is used to also train non-tethered Squirrel monkeys to move between cages for husbandry.

e. Sheltered or Outdoor Housing [*Guide*, pp. 54-55]

i. Describe the environment (e.g., barn, corral, pasture, field enclosure, flight cage, pond, or island).

There is no sheltered or outdoor housing practiced at NIDA.

ii. Describe methods used to protect animals from weather extremes, predators, and escape (windbreaks, shelters, shaded areas, areas with forced ventilation, heat

radiating structures, access to conditioned spaces, etc.).

Not applicable.

iii. Describe protective or escape mechanisms for submissive animals, how access to food and water is assured, provisions for enrichment, and efforts to group compatible animals.

Not applicable.

f. Naturalistic Environments [*Guide*, p. 55]

i. Describe types of naturalistic environments (forests, islands) and how animals are monitored for animal well-being (e.g., overall health, protection from predation).

There are no naturalistic environments used at NIDA.

ii. Describe how food, water, and shelter are provided.

Not applicable.

iii. Describe how animals are captured.

Not applicable.

C. Animal Facility Management

1. Husbandry

a. Food [*Guide*, pp. 65-67]

i. List type and source of food stuffs.

These basal diets are provided:
- High Protein Monkey Chow 5045 (LabDiet, St. Louis, MO) – NHPs
- 2018 (18% Protein) Teklad Rodent Diet (Envigo, E. Millstone, NJ) – Rodents
- Purina #5001 Rodent chow with 300 ppm fenbendazole (Research Diets, New Brunswick, NJ) – Quarantine rodents
- Rodent diet 5053 (LabDiet, St. Louis, MO) – Mice (Aponte lab)

Animal care and research programs supplement the basal diets with:
- Fruits (apple, orange, banana) – NHPs
- Vegetables (chick peas, corn, carrots, "vegetable mix") - NHPs

- Reward pellets (Bioserve) – Rodents, NHPs
- Dough diet (Love Mash®, Bioserve) – Rodent weanlings

Some rodents are feed specialized diets as the sole diet, e.g. AIN-93M diet formulated with different zinc levels (Research Diets, Inc.) for ASP 15-NRB-36; basal (D12450K), high fat (D12492) diets (Research Diets, Inc), and AIN76A modified high fat (BioServe, Inc) diets for ASP 17-INRB-18, liquid diets with varying ethanol content for ASP 17-INRB-18.

Reward pellets, e.g. nutritionally complete F0171 Rodent grain-based diet (Bioserve) are used as the sole or as supplement to the basal diet during operant testing in many rodent ASPs. Highly palatable and caloric, cafeteria diets, i.e. a mixture of human "snack foods," and also high fat rewards pellets (BioServe, Inc) are used as the sole or as supplement to the basal diet for ASPs 17-OSD-19 and 17-OSD-21, respectively.

ii. Describe feed storage facilities, noting temperature, relative humidity, and vermin control measures, and container (e.g., bag) handling practices, for each of the following:

- vendors (if more than one source, describe each)
- centralized or bulk food storage facilities if applicable
- animal facility or vivarium feed storage rooms
- storage containers within animal holding rooms

Vendors
The majority of animal diets used are shipped directly from the manufacturer, Envigo and Bioserve, to individual NIDA animal facilities and stored in designated animal facility feed storage areas without any interim vendor warehouse involvement.
Research Diets manufactured feeds are obtained (for #5001 with Fenbendazole) directly from the manufacturer or through Animal Specialties and Provisions, Inc. (ASAP, Elkridge, MD). ASAP receives palletized, shrink-wrapped bags of feed in their Redacted by agreement climate-controlled warehouse; supplemental items are purchased from local, commercial vendors. ASAP conducts a regular program of warehouse sanitation which includes monthly mopping and wall vacuuming, integrated pest management (biweekly service to inspect interior mechanical traps and exterior rodenticide/pesticide applications), and monthly sanitation of the truck dedicated to transport of separately palletized deliveries. Feed is delivered to individual animal facilities via a dedicated contractor operated truck and is stored in designated feed storage areas and freezer.

Centralized or bulk storage facilities
NIDA does not use any centralized or bulk food storage facilities.

Animal Facility or Vivarium Feed Storage rooms

Upon receipt from the vendor, animal feed is stored in rooms dedicated for food and bedding Redacted by agreement storage and food only food Redacted by agreement on separate, sanitizable plastic pallets or three-sided metal racks. (Bedding is stored in a separate, dedicated Redacted by agreement)

- The rooms are maintained below 68° F and 35-55% R.H.
- Perishable feeds are stored in walk-in Redacted by agreement and conventional Redacted by agreement refrigerator and freezers.
- Storage pallets eliminate areas for insect harborage, and a trap is present to detect room infestation. Vermin control measures are the same as described for the animal facilities, Section II.C.1.g.i., above.

Storage Containers within Animal Holding Rooms

Feedbags are transported to the housing room or behavioral testing laboratory room within the vivarium, opened, emptied into, and stored in a plastic, waterproof, vermin resistant, plastic bag-lined container.

- Each rodent housing room has a dedicated feed barrel with tight fitting lid and plastic dispensing scoop that is not moved from one room to another; plastic bag lined feed containers in behavioral testing laboratory rooms vary in size from a feed barrel to a clean, MI covered rodent cage. Separate feed barrels are dedicated for the one room of Rhesus monkeys and one shared between the two Squirrel monkey rooms.
- The housing room containers is labeled with the room number and food type. Feed is only placed in a barrel if the expiration date of the feed is greater than one month from the current date.
- The housing room feed barrel is emptied and sanitized monthly; plastic liners are changed each time a feed container becomes empty.
    - Feed barrel sanitation records are maintained in the room logbook.
    - Each feed barrel has an activity tag to record the date of filling, milling, expiration, liner change and sanitation.

Food biscuits, vegetables and fruits are prepared for NHPs in Redacted by agreement which contains a countertop surface, sink, knives and cutting boards.

iii. Describe special food preparation areas, such as feedmills and locations where special diets are formulated, if applicable. Include in the description sanitation and personnel safety practices (noting that respiratory protection is described in Section 2.I.A.2.b. ii. Standard Working Conditions and Baseline Precautions above).

Special food diets are commercially purchased (listed in C.1.a.i., above) and no special food preparation or formulation is performed on site.

iv. Describe how food is provided to various species (*ad libitum*, limited amounts, types of feeders).

Feeding Practices
Standard practice is to continuously feed rodents an excess amount (ad libitum) of biscuits. Food regulated rodents are fed daily with a prescribed number of food biscuits and or nutritionally complete, reward pellets.

Food regulated nonhuman primates are also fed daily with a prescribed number of food biscuits and or nutritionally complete, reward pellets, and a measured volume of fruit/vegetable supplements.

Food Presentation
Feed is placed into containers to minimize soiling and to facilitate access.

- Suspended V-type or wire bar feeders, integral to the cage lid or as an accessory, are used for rodents housed, respectively, in operant chambers and standard rodent cages.
- Baffled, J-type feeders are used for Squirrel and Rhesus monkeys.
- Sanitizable bowls or disposable, paper condiment cups are used for ASPs requiring placement of a daily ration of feed on the cage floor for 1) rodents that are chronically instrumented with cranial head mounts to avoid damage to cannula and hardware entrapment in feeder and cage openings, 2) veterinary support of rodents expected to have difficulty accessing overhead food due to illness, and 3) cages whose accessory holders are configured to provide two, rather than one, bottles for water-choice paradigm studies.
- Disposable, plastic petri dishes or shallow, paper condiment cups are used for dough diet for some rodent strains during weaning to solid biscuits.
- A reward pellet dispenser provides food pellets or a liquid reward dispenser provides palatable solutions during sessions if required by the experimental protocol.

v. Describe special food quality control procedures including procedures for rotating stock, monitoring milling dates, nutritional quality, bio load, chemical contaminants, etc.

Animal feed is ordered on an as needed basis as determined by facility manager's weekly inventory. The reorder level leaves an excess from the previous week's consumption on the next shipment's anticipated day of receipt; during the winter months, more feed is kept on hand in case of disruption of deliveries.

- Upon receipt, bags are inspected for mill date, punctures, tears, vermin infestation, and wetness.
  - If defects are noted, the bag is returned immediately through the deliverer or excluded from stock storage for later return.
  - The mill date is verified and any bag that is more than 90 days past milling is rejected or discarded.

- Feedbags are placed with mill dates visible in front. Feedbags are stored and used in a first in/first out manner to ensure the oldest feed is used first.
- Perishable fruits and vegetables are stored in a refrigerator or freezer used only for that purpose

b. Drinking Water [*Guide*, pp. 67-68]

i. Describe the water source, treatment or purification process, and how it is provided to the animals (e.g., bowls, bottles with sipper tubes, automatic watering, troughs, ponds, streams).

NIDA animal facilities are supplied water through municipal water system managed by the City of Baltimore.

- Municipally chlorinated water is filtered of particulates and dispensed to rodents and Squirrel and Rhesus monkeys in water bottles with sipper tubes.
- Squirrel and Rhesus monkeys in the [Redacted by] also receive chlorinated, reverse osmosis (RO) water through the Edstrom automatic watering system as a secondary water source.
- Nonhuman primate bottles are replaced with full, sanitized bottles daily.
- Sanitized rodent bottles are provided with fresh water at least once weekly concurrent with a cage change

ii. Describe methods of quality control, including monitoring for contaminants.

[Redacted by agreement] engineers test treated water to confirm free chlorine (2.0-2.5 ppm) treatment levels at the Edstrom automatic watering system's RO water bulk tank.

Water samples are collected quarterly for detection and identification of bacterial contaminants in the water bottle filler system and room faucets by Division of Veterinary Resources microbiologists and annually for chemical analysis by a commercial, certified laboratory. The City of Baltimore also provides chemical and microbiological analysis, upon request, in response to water quality concerns.

Sanitized water bottles and sipper tubes are swabbed with Sterile Ultrasnap® swabs and evaluated with a Hygiena SystemSure II™ ATPase bioluminometer monthly.

iii. If automatic water delivery systems are used, describe how they are maintained and sanitized.

Edstrom provides semiannual preventative maintenance services. Wall mounted water distribution system is automatically flushed with fresh chlorinated water daily; nonhuman primate rack manifolds and recoil hoses are drained and washed in the mechanical rack washer.

A water sample is collected from the drinking valve from racks for microbial testing to evaluate room and rack manifold sanitation.

c. Bedding and Nesting Materials [*Guide*, pp. 68-69]

i. Describe type(s) and how used for various species.

Hardwood chip bedding(7090 Teklad Sani-Chips®, Envigo, E. Millstone, NJ), heat treated by the manufacturer, is used as direct, contact bedding for rats and mice, and indirectly in excreta pans for monkeys. Hardwood chip is used as direct contact bedding and indirectly in excreta pans for rodents that are housed within operant chambers.

Compressed cotton squares (Nestlet, Ancare Corp, Bellmore, NY) for mice and crinkle paper (Enviro-dri®, Shepherd Specialty Papers, Watertown, TN) for breeding rats are used as nesting materials.

ii. Describe bulk bedding storage facilities, if applicable, including vermin control measures.

NIDA does not use any centralized or bulk food storage facilities. The bedding used is shipped directly from the manufacturer to individual NIDA animal facilities and stored in designated storage areas without any vendor interim warehouse involvement.

iii. Describe quality control procedures, including monitoring for contaminants.

In Redacted by agreement Buildings, bags are opened and emptied into automated bedding dispensers or plastic bins with a hinged lid for disbursement with a plastic scoop; empty bedding bags are discarded to the solid waste dumpster.

- Bags of bedding are inspected upon receipt for punctures, tears, vermin infestation, and wetness, and, if present, rejected or segregated for return.
- Bedding is visually examined upon opening for consistency of texture, dryness, and the presence of vermin and for freshness of aroma, and, if abnormal, segregated for review by the facility manager.

d. Miscellaneous Animal Care and Use Equipment

i. Describe motorized vehicles and other equipment (e.g., trailers) used for transporting animals, noting the type and how the cargo compartment is environmentally controlled, if applicable.

NIDA and NIH vehicles are dedicated for animal transport and provide climatically controlled cargo compartments separate from and independently controlled from the passenger compartment.

- The NIH DVR provides transportation services for animals between NIDA and other intramural programs.
- NIDA owns and maintains its own climate-controlled, animal transport van on the Bayview campus for local animal transport between NIDA animal facilities and local universities.

ii. Describe other animal care related equipment used in the animal care program (specialized equipment for exercise or enrichment, high pressure sprayers, vacuum cleaners, tractors, trailers, spreaders, etc.).

The animal facilities use HEPA filtered wet-dry vacuums, foam sprayers, mechanical floor scrubbers, and, in the Redacted by agreement cagewash, a Garb-el® waste disposal unit and in the Redacted by agreement a Nu-Star PowerTug™ (to facilitate moving heavy, castered NHP and rodent racks).

e. Sanitation [*Guide*, pp. 69-73]

i. Bedding/Substrate Change

1) Describe frequency of contact and non-contact bedding change for each species and enclosure type (solid-bottom or suspended) or pen.

The frequency to replace contact bedding, simultaneously with replacement of the soiled cage, for solid bottom caging is:

- Weekly for ventilated cages and for static, singly housed mice and rats, and,
- Twice a week for static, group housed mice and rats.
- The frequency to replace non-contact bedding is:
  - Three times a week, simultaneously with replacement of the soiled excreta pan, for NHPs, and,
  - Three times per week for rodents housed in operant chambers.

The ACUC approved cage changing intervals greater than 7 days for the centralized breeder colony ASP. Cages with a dam and new litter less than 5 days old is not changed until one week later. Hence, cage change may be delayed up to a maximum of 14 days. The delayed changing improves litter survival in sensitive strains.

2) Describe any IACUC/OB approved exceptions to frequencies recommended in the *Guide* or applicable regulations and the criteria used to justify those exceptions.

There are no exceptions to *Guide* recommended frequencies.

3) Note the location where soiled bedding is removed from the cages/enclosures and where clean bedding is placed into the cages/enclosures.

Soiled bedding

- Soiled bedding originating from centralized housing rooms is removed from cages and Squirrel monkey excreta pans into the dirty-side cagewash vacuum bedding disposal system Redacted by agreement and Garb-el® disposer Redacted by agreement, located adjacent to the tunnel washers. Rhesus monkey excreta pans are scraped clean into an MPW box and sealed within the housing room by an animal care technician wearing a fit-tested N95 respirator and face shield.
- Soiled bedding originating from behavioral testing areas is removed from test chambers and excreta pans within the testing room into plastic bag lined trashcans.
- Certain hazardous agents require bedding to be scraped and sealed in a bag within ABSL-2, PET suite animal holding room, or general laboratories' certified BSCL-2 biological safety cabinet.

Clean bedding

- Dispensed in the clean-side cagewash areas Redacted by agreement and Redacted by agreement into housing cages through the automated bedding dispensers and NHP excreta pans by hand scoop from a plastic container.
- Dispensed by hand into testing chambers and excreta pans within behavioral testing laboratories.

ii. Cleaning and Disinfection of the Micro- and Macro-Environments

Note: A description of the washing/sanitizing frequency, methods, and equipment used should be included in **Appendix 14** (Cleaning and Disinfection of the Micro- and Macro-Environment) and **Appendix 15** (Facilities and Equipment for Sanitizing Materials).

1) Describe any IACUC/OB approved exceptions to the *Guide* (or applicable regulations) recommended sanitation intervals.

There are no ACUC-approved exceptions to the *Guide* recommended sanitation intervals.

The washing/sanitizing frequency, methods, and equipment is described in **Appendix 14** (Cleaning and Disinfection of the Micro- and Macro-Environment) and **Appendix 15** (Facilities and Equipment for Sanitizing Materials).

2) Assessing the Effectiveness of Sanitation and Mechanical Washer Function

a) Describe how the effectiveness of sanitation procedures is monitored (e.g., water temperature monitoring, microbiological monitoring, visual inspections).

The mechanical operation of the cage washers and their efficacy to sanitize objects is evaluated on a regular basis.

Visually:

- Water temperature readings on the machines' touch screens are visually verified at the start of each day's operations.
- A temperature recording label (Tri-Temp® Tapes, Pharmacal Research Laboratories, Naugatuck, CT) is attached on an object prior to running the first load and attached to the log of results.
- Cages and racks are visually inspected for residual debris as they exit the washers.

Microbiologically:

- Object surfaces are sampled with sterile Ultrasnap® swabs, and swabs are evaluated for microbiological contamination with a Hygiena SystemSure II® ATPase bioluminometer.
  - Sampled surfaces include the interior of bottles, sipper tubes and other hard to reach areas.
  - Sampling is performed monthly for small objects, accessories, and standard housing equipment.

Findings that suggest inadequate sanitation are reported to the facility manager to investigate cagewasher operations, e.g. prewash and daily preventative maintenance procedures, water and steam utilities, and provision of cagewash chemicals.

The effectiveness of researcher staff-hand sanitation of behavioral testing equipment, e.g. rodent operant chambers, plastic open-field and locomotor chambers, etc, is also evaluated during the ACUC's semi-annual facility inspection. The ACUC collects samples from randomly selected chambers with Hygiena Ultrasnap bioilluminator swabs for the Facility Manager to evaluate for bacterial contamination (Hygienia SystemSure II®). The Facility Manager communicates results to the ACUC and, for results which suggest inadequate sanitation, i.e. a bioload higher than 50 Relative Light Units (RLU), the ACUC administrator informs the PI to verify staff sanitation practices and schedules additional sampling before the subsequent semi-annual inspection.

b) Describe preventive maintenance programs for mechanical washers.

Quarterly preventative maintenance services are provided to cage and rack washers in Redacted by agreement by Avant-Garde Scientific, Damascus, MD) and Redacted by agreement (by

GSH) buildings; Avant-Garde Scientific, Inc. (Damascus, MD), a commercial biomedical equipment maintenance company, is contracted to perform repairs in both buildings. All cagewash problems are repaired promptly.

Daily and weekly preventative maintenance for cage and rack washers is described in the SOP, "Soiled Side Cagewash Operations." Animal care staff:

- Clean sump screens/filters.
- Remove headers or pipe caps to manifold/arms.
- Clear jet orifices.
- Visually inspect door seal gaskets/curtains.
- Drain and flush wash and rinse tanks.
- Descale tanks and washer interior (weekly).
- Flush air or steam lines (weekly, as appropriate for machine).

f. Conventional Waste Disposal [*Guide*, pp. 73-74]

Describe the handling, storage, method and frequency of disposal, and final disposal location for each of the following:

i. Soiled bedding and refuse.

Nonhazardous laboratory and husbandry waste from the animal housing and testing rooms is collected in plastic trashcans.

- The plastic liners from the trashcans are sealed, removed, and accumulated in wheeled hoppers.
- Before the end of the day, the bags are loaded into the loading dock compacting dumpsters for weekly removal by a commercial hauler for landfill disposal.

Soiled rodent bedding is scraped into waste disposal systems within the dirty side cage wash areas:

- Redacted by agreement contains a Garb-el® waste disposal system which discharges a non-hazardous hardwood chip bedding slurry from rodent cages into the building's sanitary waste stream.
- Redacted by agreement contains a vacuum disposal system which accumulates hardwood chip bedding from nonhazardous rodent cages and NHP excreta pans to a sealed, outdoor dumpster which is emptied weekly for incineration.

Hazardous waste disposal was described in Section 2.b.iii,2),C).

ii. Animal carcasses.

For experimental animals that may die unexpectedly, carcasses are refrigerated for twenty-four hours to permit investigators to retrieve animals or to request necropsy before transfer to the freezer. Necropsy is performed on all euthanized or

spontaneously dead NHPs by board certified, DVR veterinary pathologists in Bethesda.

Animal carcasses and tissues are placed into plastic bags, sealed, and placed into cardboard MPW boxes inside a dedicated carcass freezer in Redacted by agreement Redacted by agreement and Redacted by agreement When filled, MPW boxes are sealed and stored in the dedicated cold storage room Redacted by agreement and later incinerated by the MPW contractor.

g. Pest Control [*Guide*, p. 74]

i. Describe the program for monitoring and controlling pests (insects, rodents, predators, etc.). Include a description of:

- monitoring devices and the frequency with which devices are checked
- control agent(s) used and where applied, and
- who oversees the program, monitors devices, and/or applies the agent(s).

Oversight and execution

The NIH-wide Integrated Pest Management program is managed by the DOHS, Community Health Branch, Pest Management Unit, through contracts with commercial pest control contractors to service the Redacted by agreement animal facilities. The basic concepts of the program focus on identification of pests within the facilities, use of non-chemical means (i.e. sanitation, elimination of harborage, structural exclusion of entry points, education of the animal care staff on operational practices), and, in combination with the non-chemical control measures, above, remediation through judicious application of pesticides when survey results exceed defined thresholds, e.g. an average of one cockroach per trap in two consecutive services, one mouse or rat dropping in a service area, recurring problems with other pests, e.g., flies, crickets and stored product pests which cannot be resolved using non-chemical methods , etc.

Monthly, quality control (QC) personnel surveys the facility and evaluates effectiveness of the IPM program through review of pest data and evaluates the weekly activities of the pest management technician and make/follow up on any recommendations made. In addition, a follow-up inspection is performed on previously identified pest management issues affecting the program. A report after each inspection is entered into a computer system which, after it is uploaded, is distributed to animal program supervisors and remains archived on the IPM System website.

Control agent(s) used and where applied

- The only insecticides (I) or rodenticides (R) currently approved for use are found on the IPMS web site.

Monitoring Devices

- Insect sticky traps, with surface glue adhesiveness is insufficient to immobilize a rodent, are used within all vivarium housing, testing, and support areas. Pheromone and insect light traps can be also strategically placed according to expectation of seasonal intrusions. Insect traps are monitored weekly by the pest management technician. All findings are reviewed with the Facility Manager as to the current status of the program at the completion of the weekly service.
- Live traps with transparent tops are deployed within the animal facility and monitored daily by the animal care staff for the presence of trapped research or feral rodents. Written SOPs provides instructions for biosecurity practices and humane, carbon dioxide gas euthanasia.

ii. Describe the use of natural predators (e.g., barn cats) or guard animals (e.g., dogs, donkeys) used for pest and predator control, if applicable.

Natural predators or guard animals are not used for pest and predator control.

iii. Note how animal users are informed of pesticide use and how animal users may opt out of such use in specific areas.

- Animals are not exposed to pesticides.
- All pesticide recommendations are reviewed by the facility manager and the veterinarian and approved by the Animal Program Director. If need has been determined to use pesticides inside the vivarium, animal users are provided at least one-week notification to permit evacuation of resident animals before pesticides are applied in housing or testing rooms.

h. Weekend and Holiday Animal Care [*Guide*, pp. 74-75]

i. Describe procedures for providing weekend and holiday care. Indicate who (regular animal care staff, students, part-time staff, etc.) provides and oversees care and what procedures are performed.

During weekend and holidays, four regular animal care staff members provide daily husbandry, including daily provision of food, water, enrichment, and recording of breeder colony births, veterinary care services, including animal health (e.g., morbidity/mortality) observations and treatments, and recording of environmental conditions in each animal facility.

- Supervisors meet with staff on the day prior to a weekend or holiday to review written instructions for veterinary and special husbandry care.
- Cage changing is scheduled for during the work week (Monday through Friday) to minimize weekend and holiday requirements which are limited to individual cages, as needed (spot change); rodent breeder colony are scheduled for changing seven days a week.

- Only critical facility sanitation is provided: rodent housing rooms are swept after feeding and cage changing, NHP rooms are mopped, and room trash is disposed.

ii. Indicate qualifications of weekend/holiday staff if not regular staff.

Regular husbandry staff perform weekend/holiday care.

iii. Describe procedures for contacting responsible animal care and/or veterinary personnel in case of an emergency.

Emergency contact information is posted at each entrance to the animal facilities and on the animal program web page.

- In case of a medical emergency, the Facility Veterinarian or Animal Program Director is contacted through pager/mobile phone; a rotating roster of backup emergency SciTech Services veterinarians (listed in Section I.A.2.a.i.) can be contacted when the veterinary staff is not available.
- In case of a ventilation, heating/cooling equipment, or water system failure, the animal caretaker in charge also contacts the facility manager who can muster other staff members to respond to the event, as needed.
- Additionally, the NIDA Safety, Security, and buildings' engineering groups are provided a roster of emergency contacts.

2. Population Management [*Guide*, pp. 75-77]

a. Identification

Describe animal identification methods for each species (e.g., microchips, cage/tank cards, collars, leg bands, tattoo, ear tags, brands).

- All animals are identified by printed cage card containing the vendor/source origin, strain or stock designation, PI and animal user names, arrival date, protocol (ASP) number, sex, and, environmental enrichment selection. Investigators may annotate cage cards with further identifiers such as experimental group assignment and individual identification number(s) or code.
- Individual mice and rats are identified by ear tags and notch/punches, AIMS tattoo or, for short-term identification, permanent ink markings on the tail
- Squirrel and Rhesus monkeys are individually identified by cage card, legible tattoo, and, for Squirrel monkeys, also by subcutaneous transponder.
- Toe-clipping is not practiced.

b. Breeding, Genetics, and Nomenclature

i. Describe the program for advising investigators on the selection of animals based on genetic characteristics.

NIDA has a full centralized breeding program and facility

- Every breeding need (no exception) is conducted in the breeding facility, by a team of breeder technician specialists, under the direction of a Breeder Geneticist (Technical Director of the Transgenic Facility).
- Only rodents are bred at NIDA
- Most genetically modified mouse lines have been imported from repositories or collaborators.
- Commercially available inbred and outbred rodent strains are purchased from vendors.

The Technical Director of the Transgenic Facility is the primary source for advising investigators.

- Investigators contact the Director when acquiring any new lines or developing a new project involving breeding.
- All available information is collected regarding the genetic backgrounds and histories, genotyping protocols and any other pertinent information about the lines used by NIDA investigators. The information may come from the animal vendors, literature and collaborating investigators.
- Breeding plans are designed and implemented based on the genetic information known and the requirements of the investigator.

ii. Describe the program for advising investigators on using standardized nomenclature to ensure proper reporting of the identification of the research animals with regard to both the strain and substrain or the genetic background of all animals used in a study.

- The Technical Director of the Transgenic Facility is the primary source for information on standard nomenclature for genetically modified animals. Although for practical reasons, the name of rodent lines might be simplified for internal use and communications, the official nomenclature is systematically recorded and readily available to use for official business and publications.
- Applicable guidelines such as those from the International Committee on Standardize Genetic Nomenclature for mice and the ILAR documents, "Standardized Nomenclature for Transgenic Animals" and "Definition, Nomenclature, and Conservation of Rat Strains" are used to advise investigators of proper nomenclature

iii. Describe genetic management techniques used to assess and maintain genetic variability and authenticity of breeding colonies, including recordkeeping practices (*Guide*, pp. 75-76).

Genetic management techniques

The breeding facility maintains the pedigree information (mating histories and individual animal identification) and genotype records for all the genetically modified rodents at NIDA.

Breeding plans and breeder selection are carefully chosen following these rules:

- "Wild type" rodents for breeding are systematically obtained from commercial vendors. The benefits are twofold:
  - Keep the strain background as close as possible to the vendor's or origin
  - Avoid constant inbreeding within a line and bring constantly "new blood"

  Backcrossing is done using the same strain as used at the origin (strain the line was created on or backcrossed on), unless a change of background is desired.

- Breeding plans are designed to have the minimum number of unwanted genotypes and maximum use of the offspring. When possible and appropriate, breeding plans that can produce both experimental animals and new breeders at the same time are implemented, which minimize or even can eliminate waste.
- When crossing genetically modified animals together, care is taken to avoid breeding closely related (i.e. brother-sister) animals together.
- To avoid inbreeding and limit the impact of genetic drift, most lines are going to be maintained using several lineages (origin) in parallel. Each lineage is closely monitored for genotype and phenotype at each generation.
- Lines, genotypes or backgrounds that are unique at NIDA or were difficult to produce (i.e. lengthy breeding scheme) are systematically cryopreserved.

<u>Genotyping</u>

- Genotyping is essentially all outsourced. The Technical Director of the Transgenic Facility is in charge of finding genotyping services with the most efficient outcome. Fast genotyping turnaround and high accuracy are the two main criteria allowing minimal waste of genetically modified animals. Investigators are highly encouraged to use a genotyping service having both criteria in good standing reputation and to follow the Technical Director of the Transgenic Facility advice on that matter.
- It is recommended to genotype systematically all the genetically modified rodents at least until a stable genotype can be established.
- Animals born from a breeding from which the genotype is expected (i.e. Hom x WT, Hom x Hom) are genotyped as follow:
  - All the animals from the first generation are genotyped to confirm the establishment of homozygosity.
  - At least one animal per weanling cage is genotyped for subsequent generations to keep an eye on the correct expected genotypes.
- Investigators are advised to genotype again systematically all the animals used in an experiment at the end of the experiment to confirm the genotypes of the animals used for publication.
- Newly in house created lines are genotyped with great care
  - To follow the number of copies and site of insertions (when appropriate) over time/generations.
  - Tissue samples of all breeders of newly created lines are collected for archiving at the termination of the breeding pair. These samples can be used later on to confirm genotypes or to follow potential genetic variations over several generations.

Recordkeeping practices

- Rodent breeder colony dams, sires, and offspring are identified by an additional cage card. Breeder colony cards contain the animals' generation, sire-dam pair number, sire and or dam number, dates of birth and litter size, and individual identification and genotyping information.
- Record keeping of the genotypes, strain background and pedigree of each animal are kept in:
  - Breeding colony database
  - Genotype data (in house files and commercial vendors web-sites/accounts)
  - Cage cards
  - Reconciliation breeding colony lists (systematic room census done bi-monthly in average)
  - E-mails

iv. For newly generated genotypes, describe how animals are monitored to detect phenotypes that may negatively impact health and well-being. Note that the methods used to report unexpected phenotypes to the IACUC/OB should be described in section 2.1.B.1.c.ii, "Unexpected Outcomes that Affect Animal Well-Being."

When any unexpected morbidity, morbundity, or mortality is reported to or identified by the Facility veterinarian, the veterinarian investigates the situation, contacts the investigator, and reports the situation to the APD. The APD consults with the investigator and informs the ACUC Chair.

Within the breeding colony, unexpected morbidity, morbundity, or mortality are identified by the dedicated breeding staff and first reported to the Facility veterinarian for clinical follow-up and the breeding colony manager. The manager monitors the reproductive performance of rodent breeders and correlates offsprings' animal health to identify unanticipated phenotypes that negatively impact well-being.

Additionally, researchers conduct genetic, neurochemical, and behavioral studies to characterize novel lines and detect unexpected phenotypes which may have subclinical impact on animal health and welfare.

III. Veterinary Care [*Guide*, pp. 105-132]

Note: Complete each section, including, where applicable, procedures performed in farm settings, field studies, aquatic environments, etc.

A. Animal Procurement and Transportation [*Guide*, pp. 106-109; *Ag Guide*, pp. 8; 45; 50-57]

1. Animal Procurement

Describe the method for evaluating the quality of animals supplied to the institution (from commercial vendors, other institutions, etc.).

The NIH DVR Animal Ordering and Contracts Unit and Chief of Primate Services procure rodents and non-human primates, respectively, for NIDA through established vendor contracts that define health status in terms of pathogens that are monitored and the method and frequency of monitoring.

- The vendors provide detailed descriptions of their disease monitoring and control programs to assure maintenance of acceptable health status, and NIH DVR monitors the quality assurance program of approved vendors.
- The contracts require each rodent vendor to immediately report any health status changes to the contract administrators, who in turn notify NIDA of potentially contaminated past and pending shipments. Vendor health surveillance data is supplemented by the NIDA rodent health surveillance program.

Import of animals from intramural, academic, and commercial sources not specified by contract must be approved by the NIDA APD.

- A health history of the source's animal facility is researched; supportive documentation of serological, parasitological, or other appropriate evaluation, e.g. TST, which reflects current status of the import animal's colony is reviewed; discussions, which can include colony size, location, husbandry techniques, health surveillance program, special husbandry practices, genotype information, requirement for further pre-importation testing and or prophylactic anthelmintic treatment, are held, and if believed to be pathogen-free, importation is approved.
  - Imported rodents are further evaluated by in-house quarantine to validate the source's disease-free status based on molecular (PCR) evaluation of feces and body swabs, and when indicated, serological characterization before release to research or breeder colonies.
  - Requests to import rodents from sources with unacceptable health status are rejected, or, commercial rederivation is required with review and approval of health surveillance data of the rederived offspring before importation.

2. Transportation of Animals

Describe how animals are transported between outside sources and the institution and within the institution, including loading, unloading, level of biosecurity, immune status and specific pathogen status (consider all species, including aquatic and semi-aquatic species).

Between NIDA and extramural/intramural institutions

- Approved vendors and intramural sources deliver animals directly to facilities via, respectively, vendor or NIH DVR operated, climate-controlled vehicles.
- Transportation of non-contract rodents between NIDA and extramural institutions is portal to portal via commercial animal transporters and local transport after obtaining appropriate clearances.

- Laboratory administrators purchase shipping services through a commercial animal transporter, commonly World Courier (New Hyde Park, NY), to facilitate compliance with animal transportation requirements including provision of health certificates, sending/receiving institutions' addresses, and husbandry and veterinary care contacts. The animal program staff also transports rodents via the temperature controlled, NIDA animal transport vehicle between NIDA and collaborators at local universities', e.g. the Johns Hopkins University and University of Maryland, animal facilities.
- The animal facility manager serves as shipping coordinator for packing up rodents for export in disposable, plastic or cardboard, filter-protected shipping containers (rodents) with sufficient internal food and water sources and for receiving imported rodents.

• Additional animal biosecurity measures include:
  - Animals are delivered to a dedicated Redacted by agreement NIDA animal facility and Redacted by agreement general loading docks in disposable, filter-protected shipping containers (rodents) and, after completion of NIH quarantine in Poolesville, Maryland, in metal or disposable wooden shipping crates (Rhesus and Squirrel monkeys) only to the Redacted by agreeme dock.
  - Crates are transferred to a cart for transport directly to their designated housing room, and the exterior surfaces of rodent shipping crates are spray misted in-transit with chlorine dioxide solution prior to unpacking and transfer to their home cages.

Between and within NIDA sites

• The animal care staff transports rodents for investigators between the Redacted by agreement and Redacted by agreement buildings.
• Transportation of animals between housing areas and laboratories, whether inside or outside the animal facility, occurs within covered cages or containers.
  - Rodents are transported within individual cages covered by a microisolator (MI) lid between housing and procedure rooms and via a freight elevator within the same building and via the temperature controlled, NIDA animal transport vehicle between Redacted by agreement animal facilities.
  - Squirrel monkeys are individually transported within a stainless steel transport box, which has a hinged, locking perforated lid and handle, within the primate corridor of the animal facility to reach behavioral testing, fMRI, or PET suite rooms.
  - Conscious Rhesus monkeys are transported between housing room and adjacent testing room through the isolated NHP corridor while secured within a castered restraint chair. Anesthetized Rhesus monkeys are transported through vivarium corridors on a covered cart between housing room and NHP surgery suite through the isolated NHP corridor suite or within a castered stainless steel transport container thorough the vivarium corridor to the PET suite and further, the adjacent corridor to reach the clinical MRI suite.

B. Preventive Medicine

1. Animal Biosecurity [*Guide*, pp. 109-110]

a. Describe methods used to monitor for known or unknown infectious agents. Note that if sentinel animals are used, specific information regarding that program is to be provided below.

Ongoing and episodic systems evaluate animals' health status for known or unknown infectious agents:

- The commercial vendors' surveillance and monitoring program is incorporated into NIDA's own monitoring program, to prevent introduction of murine pathogens.
- A sentinel surveillance program and daily animal observations by trained animal care technicians monitor for infectious agents:
  - Sprague-Dawley rats and Swiss Webster mice are purchased from a commercial vendor and are continuously used as sentinel animals for each rack in each centralized rat and mouse housing room. Sentinels are exposed to bedding used previously by other animals on that rack.
  - Sentinel rodents are evaluated serologically and replaced on a semiannual frequency; molecular (fecal and fur swab PCR) testing, at quarterly intervals, is also performed on the sentinel rodents.
- Pest control practices are applied building-wide, and live-traps are continually deployed and monitored to detect pest and vermin infestation within the animal facilities which can support evaluation of health status of feral rodents. SOP directs elimination of escaped research and feral animals; risk-based testing of feral rodents is limited to animals detected within an animal housing/testing room.
- As need arises, imported nonhuman primates and non-commercial origin rodents are tested during entry quarantine, and biologic materials destined for administration into animals, identified in the ASP's Section L, Biological Material/Animal Products, may undergo PCR or Mouse Antibody Production (MAP)/Rat Antibody Production (RAP) testing to ensure the products are free from zoonotic and other rodent agents.

b. Describe methods used to control, contain, or eliminate infectious agents.

Control

- Only animals of a desired health status enter housing and testing rooms by management of procurement from commercial vendors and by quarantine evaluation of non-commercial sources.
  - In addition to parasitological evaluation, all imported rodents are prophylactically treated with anthelmintic drugs to reduce risks from undetected parasites

- Integrated Pest Management practices approach focus on excluding entry of feral rodents.
- Animal care staff is precluded from owning pet rats and mice which may serve as a source for contamination; research staff is queried and educated during orientation of risk from pet rodents and from contact with animals at collaborating institutions' animal facilities. Building management staff is additionally briefed on contamination risks from vermin and feral pests and from non-biosecure building areas, e.g. exterior dumpster.

Containment

- Maintenance of a centralized animal facility further controls potential for cross-contamination of infectious agents:
  - Cage wash provide clearly defined areas for clean and soiled equipment and operations.
  - Assignment of animal care personnel, traffic control patterns, and physical segregation contain risks between populations (e.g. breeder colony and experimental rodent housing rooms) within and between defined, conventional housing and potentially infectious, quarantine and ABSL-2 housing areas.
  - Use of designated PPE, ventilated caging systems, laminar flow change stations, transport of rodents within microisolator covered cages, and negative room differential air pressurization aid in agent containment.
- These practices and procedures ensure appropriate sanitation of supplies, equipment, and facility reduce potential for cross-contamination:
  - Disinfectants are applied to the exterior of rodent shipping crates upon arrival.
  - Potential for fomite transmission of infectious agents is reduced by sanitation practices, defined in each species specific SOP, Section 16: Sanitation of Testing Equipment and Rooms, for animal testing equipment.

Elimination

- The method for elimination of the infectious agent depends upon the agent, population of animals infected, and purpose of the study. Elimination may involve isolation, treatment, or euthanasia of animals which may be done in a cohort, by rack, by room, or the entire facility depending on the nature of the infectious agent. Common methods employed include for:
  - Mites - treatment with topical selamectin.
  - Pinworms- treatment with fenbendazole medicated diet
  - Viral agents: test and cull for some agents, or "burn-out" by temporary cessation of importations and breeding.

2. Quarantine and Stabilization [*Guide*, pp. 110-111]

a. Describe the initial animal evaluation procedures for each species.

Mice and Rats

- Animal care staff first evaluate the integrity of the rodent shipping container upon receipt at the Redacted by agreement vivarium loading docks to infer biosecurity risks from the delivered animals. Infrequently, intramural or university originating rodents are directly delivered, following ACUC approved ASP, to general laboratories for immediate terminal use.
- During uncrating and initial caging within research and quarantine housing rooms, animal caretakers visually evaluate all rodents for overt signs of disease or physically abnormal animals.
- The facility manager, animal health technician, and/or veterinarian are notified of evaluation findings to obtain instruction and or clinical care.

Squirrel and Rhesus monkeys

- After completing an NIH entry quarantine at the National Institutes of Health Animal Center (Poolesville, MD), the veterinarian and/or animal health technician visually inspects each animal as it is unloaded, at the Redacted by agreement vivarium loading dock, and after uncrating into its individual cage.
- Accompanying medical records are reviewed by the veterinarian, and any clinically ill or injured animals are examined and treated by the veterinarian.
- After stabilization, NHPs undergo a pre-study health assessment that includes a physical examination, dental hygiene and exam, a complete blood cell count (CBC) and serum chemistry profile, and an electrocardiogram (ECG). An identification transponder is subcutaneously implanted in Squirrel monkeys and, as needed, tattoo reapplied to NHPs

b. Describe quarantine facilities and procedures for each species. For each species, indicate whether these practices are used for purpose-bred animals, random-source animals, or both.

Facilities

Isolation and quarantine for rats and mice is performed within a suite of rooms in the Redacted by agreement vivarium which provides negative differential pressures to adjacent areas. An anteroom (Redacted by agreement permits staging and assembly of clean cages, assembly of anthelmintic treatment and diagnostic sample collection materials, and donning PPE before entry to the quarantine room. The quarantine room, Redacted by contains a laminar flow hood change station and Redacted by agreement for housing. The contiguous corridor contains two-side autoclaves for material pass-through and an anteroom for personnel and rolling stock (racks, carts) to exit to the dirty side Cage Wash Redacted by agreement

NIH Policy (PM 3044-1: Nonhuman Primate Quarantine) requires NHP quarantine at the Poolesville, MD, NIH Animal Center, a fully AAALAC accredited NIH animal facility, for all NHPs originating from extramural sources.

Procedures

Quarantine of both purpose bred (rats, mice, rhesus and the great majority of squirrel monkeys) and random source (a minority of the squirrel monkey colony) species are handled identically.

Rats and Mice

Rodents originating from non-approved vendors and or non-commercial sources are housed in the quarantine suite until release to animal housing rooms.

- Pooled fecal samples from each cage and fur swabs from each individual animal is collected for molecular (PCR) screening for endoparasites and ectoparasites.
- Based on review and evaluation of the health status and history of the originating institution, pooled fecal samples from each cage is evaluated through molecular (PCR) screening for an abbreviated panel of agents or a blood sample from each individual animal is serologically tested for an expanded panel of viruses.
- Unless precluded by scientific considerations, all rodents receive prophylactic anthelmintic treatment for endoparasites (300 PPM fenbendazole medicated rodent food) and ectoparasites (12 mg/kg, selamectin, topical).

If animals test positive for pathogens that would preclude entry into research and breeding colonies, they can be commercially rederived, treated, held for passive elimination of disease, tested in isolation, or euthanized

Nonhuman Primates

Briefly, quarantine procedures at the NIH Animal Center include:

- Vendors screen animals to define NHPs and provide complete copies of all procured animals' health records, including the origin and date of birth, if known.
    - At least one negative TST must be documented within 30 days prior to shipment.
    - Macaques species are polymerase chain reaction (PCR) negative and seronegative for Simian Retrovirus (SRV), herpes B virus (Cercopithecine herpesvirus 1), Simian Immunodeficiency Virus (SIV) and Simian T-Cell Leukemia Virus (STLV-1).
- NHPs that have resided in domestic colonies for more than one year prior to arrival at NIH undergo a standard sixty (60) day long quarantine and is extended to ninety (90) days where any part of the cohort has arrived in the U.S. within one calendar year of arrival at NIH.

Squirrel monkeys

- Squirrel monkeys complete three consecutive negative tuberculosis skin tests at two-week intervals, hematological screening (CBC and serum chemistry profile), parasitological examinations, viral profile testing (Herpesvirus tamarinus, Herpesvirus saimiri, SquiCMV, Rubeola), and, when requested, vaccination (e.g. Rubeola).

Rhesus monkeys

- Macaque species complete four consecutive negative tuberculosis skin tests at two-week intervals, hematological screening (CBC and serum chemistry profile), parasitological examinations, viral profile testing (SIV, SRV 1,2,3,5, STLV-1,2, Cercopithecine herpesvirus 1, Rubeola), and, when requested, vaccination (e.g. Rubeola).

For NHPs demonstrating suspect or positive reactions on tuberculosis skin testing, or determined to be infected with tuberculosis, the NIH Animal Center follows defined procedures (Guidelines for the Prevention and Control of Tuberculosis in Nonhuman Primates in the NIH Intramural Program) for monitoring, husbandry practices and procedures, and continued testing of cohorts. Positive tuberculosis reactors are precluded from the NIDA facility.

c. Describe the required/recommended stabilization period for each species.

After receipt, rodents are provided at least 72 hours of acclimation before experimental use unless exempted by ACUC-approved ASP, e.g. time-sensitive tissue harvest procedures.

Primates are provided a minimum of 7 days for acclimation before use unless exempted by ACUC-approved ASP, e.g. transient animals used in collaborative PET or MRI imaging studies.

3. Separation by Health Status and Species [*Guide*, pp. 111-112]

a. Describe the program for the separation of animals by species, source, and health status. If the animals in different status are not maintained separately, describe circumstances in which mixing occurs and explain the rationale for mixing.

Animals are separated by species and health status. Animals of the same species and health status, but from different sources, may be housed in the same room since, in many cases, animals belong to the same investigator.

b. Describe situations where multiple species may be housed in the same room, area, or enclosure.

Multiple species, or the same species obtained from different sources, can be housed within separate cubicles within Redacted by agreement for quarantine Redacted by agreement and for ABSL-2 containment Redacted by agreement

Rats and mice have been housed in separate ventilated cage racks within a Redacted by agreement Redacted by agreement to increase space efficiency after notification of and approval from all of room's principal investigators.

Multiple species are not housed within the same enclosure.

c. Describe isolation procedures and related facilities for animals.

In most cases, ill animals are treated and remain in their home cage.

Clinically ill rats and mice can be housed overnight within separate intensive care units in the Redacted by agreement animal treatment room, Redacted by agreement The quarantine suite cubicles can be used to hold transient NHPs involved in collaborative imaging studies, and, when indicated by risk assessment, resident NHPs or rodents can be housed in a cubicle for observation and or treatment.

C. Clinical Care and Management [*Guide*, pp. 112-115]

1. Surveillance, Diagnosis, Treatment and Control of Disease [*Guide*, pp. 112-113]

a. Describe the procedure(s) for daily observation of animals for illness or abnormal behavior, including:

- the observers' training for this responsibility
- method(s) for reporting observations (written or verbal)
- method(s) for ensuring that reported cases are appropriately managed in a timely manner.

Animal care staff, who are trained to recognize signs of disease and abnormal behavior of the animals they work with, make cage side observations twice daily, Monday through Friday, and once daily on weekends and holidays. They report ill and other abnormal animals to the veterinary care staff for timely examination, triage, and treatment.

- The veterinarian and facility supervisors provide animal caretakers with on-the-job training during staff meetings or training sessions, AAALAS certification training, and while performing their other duties.
- Animal caretakers report abnormal observations on the written Daily Room Sheet for each housing room to the supervisor and/or facility manager who, in turn, informs the facility veterinarian, animal health technicians, and APD via a twice-daily electronic mail report. If pain or distress is present, the animal caretaker, weekend supervisor, or facility manager, also verbally notifies the facility veterinarian or animal health technicians immediately.
- After hours, including weekends and holidays, the animal care and research staff contacts the veterinarians and facility supervisors through personal electronic communicators (mobile phones, pagers, etc). The animal care staff additionally contacts the on-call veterinarians or animal health technician to discuss triage and treatment of newly emerging or previously identified abnormal animals.
- A roster of animals that were found dead is also maintained for each housing room and electronically reported to the Veterinary Care and appropriate research staff.

Many of the animals involved in behavioral research are also observed daily during testing sessions and in laboratory housing areas by researchers, who are able to note subtle behavioral or body weight changes. Methods of communication between research staff and animal/veterinary care staff is described directly below.

b. Describe methods of communication between the animal care staff and veterinary staff and the researcher(s) regarding ill animals.

Research Staff

- Posted emergency signs provides research and program support staff with contact information for responsible animal care and/or veterinary personnel.
- The research staff communicates to the animal/veterinary care staff directly by telephone or electronic mail, by verbally informing the animal caretaker, or by submitting a written service request through the facility manager.

Animal/Veterinary Care Staff

- The animal care staff communicates animal health issues to veterinary care staff via a daily electronic mail report, and also, verbal notification for animals in pain or distress.
- The animal care staff also directly notifies research staff of animal mortality through a File Maker database generated, electronic mail report.
- The veterinary care staff communicates animal health issues to researchers in a File Maker database generated animal health report, progress of on-going treatments, and inquiries of the investigators' plans for the disposition of the affected animal through electronic mail, and they communicate verbally by telephone to clarify recent research use and, for moribund animals, to rapidly schedule euthanasia.
- Emergency contact information for Principal Investigators, or responsible designees, for both during and after work hours, is collected for each ASP and maintained in binders which are accessible to the animal and veterinary care staff for contact through telephone to discuss animal condition and disposition.

After hours, including weekends and holidays, animal caretakers and research staff verbally contact the veterinarians, animal health technicians, facility manager through personal electronic communicators.

c. Describe the preventive medicine and health management/monitoring programs (e.g., physical examination, TB testing, vaccination, hoof/nail trimming, teeth cleaning/floating, vendor surveillance, use of sentinel animals) for each species.

Nonhuman primates (NHPs)

As described above (Section B.2.a.), the preventative medicine and health programs starts during quarantine at the NIH Animal Center (Poolesville, MD) and, continues with animals' initial evaluation, a pre-study health assessment, soon after receipt at NIDA.

The preventative medicine program for resident NHPs is risk adjusted for animals which actively participate in research studies and animals which are not actively used or off-study.

- Tuberculin skin testing is performed semiannually (Squirrel monkey) and quarterly (Rhesus monkey) along with body weight measurement, confirmation of identification, and dental care, as needed, for all NHPs
- During active study participation, NHPs receive quarterly health examinations including dental exam and care, as needed, complete blood cell (CBC) count or hematocrit, serum chemistry profile, and for Squirrel monkeys, annual indirect blood pressure and electrocardiography (ECG) screening. Many NHPs on active study are weighed daily as part of their experimental use.
- NHPs that are held off-study receive at the minimum a semi-annual health assessment and an annual CBC count or hematocrit, serum chemistry profile, and for Squirrel monkeys, annual indirect blood pressure and ECG screening.

NHPs that die unexpectedly, or are euthanized, are submitted for necropsy to the DVR especially for tuberculosis evaluation and disease surveillance.

Rodents

A sentinel surveillance program and daily animal observations by trained animal care technicians monitor the health of resident rats and mice.

- Sprague-Dawley rats and Swiss Webster mice are purchased from a commercial vendor and are continuously used as sentinel animals for each rack in each rat and mouse housing room.
  - Each rat or mouse housing rack is sentinelized with one cage containing two Sprague Dawely rats or three Swiss Webster mice, respectively.
  - Sentinels are exposed to bedding used previously by other animals on that rack.
- During scheduled cage changes, soiled bedding samples are pooled from the one side of each rat and mouse rack and added to their sentinel's cage.
- Blood is collected from sentinel rodents every 6 months for serological testing and sentinel fecal pellets and principal rodent fur swabs are collected four times per year between the serological testing samplings for the presence of these agents:

Mouse Agents	4/year	Twice yearly
Mouse Rota Virus (EDIM)	X	X
Minute Virus of Mice (MVM)	X	X
Mouse Hepatitis Virus (MHV)	X	X
Mouse Norovirus (MNV)	X	X
Mouse Parvovirus (MPV)	X	X
Mouse Theilovirus (TMEV-GDVII)	X	X
Mycoplasma pulmonis		X
Pneumonia Virus of Mice (PVM)		X
Respiratory Enteric Orphan Virus (Reovirus)		X

Sendai virus		X
Ectromelia virus		X
Lymphchoriomeningitis virus (LCMV)		X
Rat Agents		
Kilham Rat Virus (KRV)	X	X
Rat Corona Virus (RCV/SDAV)	X	X
Rat Parvovirus (RPV)	X	X
Rat Theilovirus (RTV)	X	X
Toolan's H-1 Virus (H-1)	X	X
Parvovirus (PARVO)		X
Rat Minute Virus (RMV)		X
Mycoplasma pulmonis		X
Pneumonia Virus of Mice (PVM)		X
Respiratory Enteric Orphan Virus (Reovirus)		X
Sendai Virus		X
Pneumocystis carinii (Rat Respiratory Fungus)		X
Lymphchoriomeningitis virus (LCMV)		X

- After receipt of serological testing results, rodent sentinels are culled. Organs are grossly examined for lesions, DVR pathologists perform histopathology on any tissues with gross lesions

2. Emergency Care [*Guide*, p. 114]

a. Describe the procedures to ensure that emergency veterinary care is continuously available for animals during and outside of regular work hours, including access to drugs or other therapeutics and equipment.

Veterinary care is continuously available.

- The full-time, APD and facility veterinarian are available during and outside of regular working hours and coordinate planned absences.
- Emergency veterinary care coverage on weekends and holidays is provided by on-call laboratory animal trained veterinarians by contract arrangement.
- Telephone contact information to reach a veterinarian is posted in both animal facilities and on the animal program's web page.

Treatment supplies and equipment are accessible.

- Therapeutic drugs are stocked in the vivarium Redacted by agreement
- Controlled substances including anesthetic and euthanasia drugs are secured and accessible in the treatment room.
- Anesthesia equipment, surgical instruments, and clinical supplies are readily available in the treatment room and surgical suite.

b. Describe the authority of the Attending Veterinarian or his/her designee relative to the emergency treatment of animals in the program.

The APD, acting as the Attending Veterinarian, delegates the authority to the facility veterinarian and backup emergency veterinarian to provide emergency care including treatment for illness and pain, suspension from use in a study, and, when necessary, euthanasia. Although the veterinarians attempt to accommodate both scientific as well as animal welfare objectives, e.g. rapidly schedule terminal perfusion or collect tissues after euthanasia, the veterinarians have the authority to appropriately treat any animal in a timely manner to relieve potential pain or distress.

3. Clinical Record Keeping [*Guide*, p. 115]

a. Describe the procedure for maintaining medical records and documenting treatment of ill animals including: clinical laboratory findings, diagnoses, treatments, medical progress records, etc. Identify the species for which individual records are maintained and where such records are kept.

Comprehensive individual medical records are maintained for nonhuman primates. Individual medical records for mice and rats are limited to experimental and therapeutic surgery and to clinical care of ill rodents.

Medical Record Documentation Practices

- The rodent surgical record, e.g. date, anesthetic drug dosage, surgery type and location/stereotaxic coordinates, and surgeon name/initials are recorded within laboratory notebooks
- Postsurgical rodent records identify the animal, date and type of surgery, and documents provision of ASP specified treatments, e.g. postoperative analgesia, and or observations of animal activity and surgical wounds.
- The Facility Veterinarian and the animal health technician maintain permanent medical records of preventative, clinical veterinary, and post-surgical care for individual nonhuman primates and printed animal health reports for individually identified rodents that are currently receiving treatment or being monitored.
    - Permanent, written NHP records are created upon arrival at NIH and assimilate all available information from the animal importer or breeding colony, records from quarantine(s), and other intramural institutes.
        - Written entries are added to the chronological medical record to summarize ancillary clinical pathology, microbiology, parasitology, and pathology findings
        - Surgical records, anesthesia records, and physical exam forms, are included as hard copy attachments.
        - Upon resolution of a case, the NHP individual medical record is returned to its file cabinet.

- All rodent clinical cases are entered into a File Maker database system to create the initial medical record,
  - The database generates an animal health report which is sent by electronic mail to researchers and a printed hard copy is placed within a binder, sorted by room number, for written annotation with progress notes, documentation of treatments, and resolution.
  - Researchers are informed of progress and prognosis through verbal communication and electronic mail.
  - Upon resolution, the File Maker system is updated reflect resolved status and annotated hard copy is transferred to a file cabinet from its binder for archiving.

Record Maintenance

The following section (b.) addresses where the records are kept/maintained.

b. Identify individual(s) (titles, not necessarily names) responsible for maintaining such records and identify where the records are maintained and who, including the IACUC/OB has access to the records.

Record Maintenance

- Each PI's ASP identifies the researcher(s) responsible for anesthesia, surgery, and post-surgical records for mice and rats. Laboratory notebooks containing the surgical records are kept within the animals' laboratory. The rodent postsurgical care record is maintained in the animal's cage card holder for the acute period of postoperative observation, and investigators may later archive the record, typically within the animals' laboratory.
- The Facility Veterinarian and animal health technicians maintain permanent medical records of preventative, clinical veterinary, and post-surgical care for individual nonhuman primates and printed animal health reports for mice and rat in, respectively, Redacted by agreement and Redacted by agreement

Records' Access

- The veterinarians (including the Attending Veterinarian), animal health technician, and ACUC have access to medical records; clinical records are made available to investigators if needed.

c. Describe the role of the Attending Veterinarian in recordkeeping.

The APD, acting as the Attending Veterinarian, delegates the authority to the facility veterinarian and backup emergency veterinarian to provide veterinary care and its accompanying responsibility to maintain medical records. The APD, independently and as a member of the ACUC, maintains oversight responsibility for institutional recordkeeping.

4. **Diagnostic Resources.** Describe available diagnostic methods used in the program including:

a. In-house diagnostic laboratory capabilities.

Redacted by agreement contains in-house diagnostic resources. Equipment includes a light microscope and dissection scope, sink for bacterial or cytological staining, centrifuges for blood tube and microhematocrit tube separation of serum, parasitological floatation supplies, and specimen refrigerator.

In addition, the intramural DVR's diagnostic laboratories perform environmental and clinical bacteriology, parasitology, diagnostic PCR testing, and necropsy of NHP carcasses.

b. Commercially provided diagnostic laboratory services.

A full-service commercial clinical laboratory, IDEXX laboratories, picks up samples and performs the majority of the clinical pathology (hematology and serum chemistry) testing, clinical bacteriology, and, through the NIH DVR diagnostic laboratories contract, performs rodent pathogen serological and molecular diagnostic testing.

c. Necropsy facilities and histopathology capabilities.

The veterinary care staff utilizes the same resources as are provided to research staff for euthanasia and processing of carcasses for tissue collection: Dedicated, common use necropsy areas are located in Redacted by agreement and Redacted by agreement which contain downdraft stainless steel necropsy tables with sinks, MPW boxes, and a carcass freezer.

Although many research laboratories perform histological and histopathologic evaluation of rodent brain tissues after terminal perfusion, the intramural DVR's diagnostic laboratories also provides clinical necropsy and histopathological services by board-certified veterinary pathologists at their Bethesda, MD, facilities.

d. Radiology and other imaging capabilities.

A 30MA portable machine (Min-X-Ray®), protective equipment (vest, gloves, neck collar), automatic film developing machine and a film vault with cassettes containing rare earth intensifying screens, and an ultrasound machine (Aloka SSD-1400) with 7.5MHz probe is maintained in the Redacted by agreement suite.

5. **Drug Storage and Control**

a. Describe the purchase and storage of controlled and non-controlled drugs.

DEA Schedule I through V drugs are regularly studied and used in animal studies. The Chief Pharmacist of the NIDA Intramural Research Program oversees all acquisition, storage, and distribution of controlled substances.

- All schedule drugs are ordered, received, recorded on inventory, and stored by the NIDA IRP pharmacy. The pharmacy dispenses drugs and records the transfer to an authorized recipient.
- Small quantities that are for relatively immediate use by nonclinical laboratories are maintained in primary storage containers within secondary containment (e.g. locked doors, lockers, combination safes, etc.) in a designated area. Access is controlled by limiting and assigning keys and card access to individuals.
- Some experiments may necessitate that solutions of controlled substances be kept in the procedure rooms for extended periods, e.g. in a syringe driver or pump for automated, chronic or repeated programmed infusions to animals at prescribed times. These controlled substances solutions are prepared at low concentrations that are exempt from the DEA schedule.

Non-controlled drugs are ordered from commercial vendors and received by the ordering investigator. A supply of therapeutic veterinary drugs is stored in cabinets in animal treatment areas, Redacted by agreement for use and dispensing by the Veterinary Care staff.

- The expiration dates of drugs and expirable supplies highlighted or marked to facilitate monthly review of stock.
- Each item is stored and used in a first-in-first-out manner. Items that are identified as being within one month of expiration date, replacement ordered, and discarded after the next month.

b. Describe record keeping procedures for controlled substances.

The authorized recipient of controlled substances maintains an inventory which includes:

- Name of controlled substance,
- Unit size received,
- Amount received,
- Date used,
- Signature of user, and
- Amount on hand.

The Pharmacy conducts regular audits of recipients' inventory of control substances to reconcile inventory with usage records.

D. Surgery [*Guide*, pp. 115-123]

1. Pre-Surgical Planning [*Guide*, p. 116]

Describe the process(es) used to ensure adequate pre-surgical planning, including: identifying personnel; locating equipment, supplies, veterinary involvement for selecting analgesic and anesthetic agents and facilities; planning; and pre- and post-operative care.

In the ASP, the Principal Investigator (PI) -
Identifies:
- Surgeon(s) and responsible postoperative care provider(s), and
- Location for the surgical procedure

Describes:
- Preoperative preparation,
- Anesthetic and analgesic use,
- Surgical procedure method,
- Postoperative care, health monitoring and record keeping.

Assures:
- Personnel performing surgical procedures are qualified through experience, or that
- Personnel will be trained to perform the surgical procedures described in the ASP.

To ensure adequate pre-surgical planning,
- The NIDA ACUC provides investigators with documents, "Anesthesia Guidelines for Rodents" and "Analgesia Guidelines," which are regularly updated by the APD to reflect contemporary, best practices, to determine the need for analgesia and choice of analgesic drugs for the most commonly performed surgeries.
- Species specific Standard Operating Procedures documents describe preoperative and postoperative procedures.
- The APD provides veterinary guidance for selecting analgesic and anesthetic agents regarding specific applications during ASP preparation by consultation with the investigator and during subsequent ACUC review of potentially painful procedures including surgery.

2. Surgical Facilities [*Guide*, pp. 116-117, 144-145]

List building name(s) and room number(s) or other locations (coded, if confidential) where surgical procedures are performed. For each, describe:

- the type of species (including rodents, fish, agricultural species, etc.)
- nature of procedure(s) (major/minor/emergency, survival and non-survival, etc.)
- the amount of use [heavy (daily), moderate (weekly), or light]
- major surgical support equipment available (gas anesthesia machines, respirators, surgical lights, etc.)
- facilities for aseptic surgery, surgical support, animal preparation, surgeon's scrub, operating room, and postoperative recovery
- construction features of the operating room(s), including interior surfaces, ventilation, lighting, and fixed equipment used to support surgical procedures and other means of enhancing contamination control

Note: If preferred, the information requested in this section may be provided in Table.

Non-rodent survival surgery is performed in Redacted by agreement. Surgical area usage is light; approximately 40 chronic intravascular catheter implantations (90-minute duration)

for self-administration studies or removals (40-minute duration) are performed yearly in Squirrel monkeys.

Survival and non-survival rodent surgeries occur in dedicated locations within animal laboratories and dedicated rodent surgery rooms in the Redacted by agreement numbers) buildings.

- Rodent non-survival surgeries, e.g. craniotomy and or catheterization for in vivo electrophysiology procedures, are performed at these locations:

Redacted by agreement	Major	Moderate
	Major	Moderate
	Major	Moderate
	Major	Moderate
	Major	Moderate
	Major	Moderate
	Major	Moderate
	Major	Light
	Major	Light
	Major	Light
	Major	Light
	Major	Light
	Major	Light
	Major	Light
	Minor	Light
	Minor	Light
	Major	Light

- Rat and mouse survival surgery, e.g. craniotomy procedures, are performed at these locations:

Redacted by agreement	Major	Light
	Major	Moderate
	Major	Heavy
	Major	Heavy
	Major	Moderate
	Major	Moderate
	Major	Moderate
	Major	Heavy
	Major	Moderate
	Major	Moderate
	Major	Light
	Major	Light
	Major	Light
	Major	Light
	Major	Light
	Major	Moderate
	Major	Moderate

Redacted by agreement	Major	Moderate
	Major	Moderate
	Major	Moderate
	Major	Light
	Major	Light
	Major	Light
	Major	Light
	Major	Light

Aseptic Surgery Facilities

NHP survival surgery is restricted to the animal program's centralized non-rodent survival surgery suite. The nonrodent surgery suite consists of four rooms in the Redacted by agreement

- Redacted by agreement
- Redacted by agreement Preoperative preparation and postoperative recovery).
- Redacted by agreement functions as surgeon change area. The suite's internal corridor Redacted by agreement contains a scrub sink for surgeon preparation.
- Redacted by agreement functions as the instrument preparation area. Sterilization and supply storage also occurs in this room.

Rodent surgeries occur in dedicated locations within animal laboratories and dedicated rodent surgery rooms in the Redacted by vivarium and Redacted by agreement Facility design and designation of shared space Redacted by agreement encourages investigators to designate rodent surgery areas to rooms within the vivariums. The need for ABSL-2 containment, hazardous agent use, and specialized electrophysiology equipment are also considerations for designation of surgical areas.

Construction Features

Surgical suite and general animal facility construction features are identical.

- Walls are epoxy paint finish over filled masonry block and over gypsum panels and metal frame walls.
- Floors are troweled, seamless epoxy resin coved to a height of 6 inches at the wall/floor junction.
- Ceilings are epoxy paint finish over gypsum panels with water resistant, recessed, fluorescent lighting banks.
- Doors are fiberglass or galvanized steel frames, solid or with observation windows.
- The ceiling contains four supply air diffusers along the center of the room and one exhaust air diffuser near a room corner. A ceiling column contains electrical outlets and waste anesthetic gas vacuum connection.

Major Surgical Support Equipment

Major equipment in the nonrodent surgery suite Redacted by agreement includes:

- Ceiling mounted, overhead surgery lights
- Hallowell® gas anesthetic ventilator workstation
- VSSL® adjustable, warming surgery table
- SpaceLabs® ECG monitor

- Welch Allyn Propaq® ECG/capnograph/oximeter monitor
- VetPro VIP 2000 Veterinary Infusion Pump
- Parks Medical Electronics, 811-D Ultrasonic Doppler Flow Detector
- Hazard Technology® Containment (HEPA) Vacuum
- Cardiopulmonary "crash kit" with Ambu bag
- Getinge 533LS vacuum steam sterilization autoclave
- 3M Steri-Vac™ 5XL Ethylene oxide gas sterilizer and aerator.
- Alternative perioperative warming devices (circulating warm-water warming blankets, BAIR hugger® forced-warm air blower, Lyons® animal intensive care incubator, Emerson® heating lights)

Rodent surgery areas typically contain a stereotaxic apparatus, table top or wall/ceiling mounted surgery light, HEPA filtered vacuum, and magnifying scopes for delicate surgery work.

3. **Surgical Procedures** [*Guide*, pp. 117-118]

a. Describe the criteria used to differentiate major from minor survival surgery, including classification for certain procedures (e.g., laparoscopic technique).

Major surgery is considered any surgical intervention that penetrates and exposes a body cavity; any procedure that has the potential for producing permanent physical or physiological impairment; and/or any procedure associated with orthopedics or extensive tissue dissection or transaction.

The ACUC classifies rodent craniotomies for cannulation and injection, regardless of cavity exposure, as major surgeries.

Minor surgery is considered any surgical intervention that neither penetrates and exposes a body cavity nor produces permanent impairment of physical or physiologic function. The ACUC classifies chronic vascular catheterization as minor surgery; external iliac vein catheterization, performed through a retroperitoneal approach in Squirrel monkeys, is considered major surgery if the body cavity was inadvertently penetrated.

b. How is non-survival surgery defined?

Non-survival surgery is a surgical procedure performed on an animal that is euthanized at the end of the surgical procedure while still under surgical anesthesia.

Transcardial perfusion is defined as a euthanasia procedure rather than non-survival surgery.

4. **Aseptic Technique** [*Guide*, pp. 118-119]

a. Describe procedures, equipment, and protective clothing used for aseptic surgery. Include patient and surgeon preparation.

After induction of anesthesia, hair is removed from the surgery site with clippers (or, for mice, fur plucking), the animal is positioned for surgery, and the skin incision site is disinfected by scrubbing with (iodophor or chlorohexadine-based) surgical soap followed by alcohol or sterile water rinse, repeated three times in an inside-out spiral pattern. The surgery site may then be protected with a sterile pad if further positioning is required for surgery. For non-human primate surgery, a large sterile drape is placed over part of the table and animal, a sterile transparent, fenestrated drape is placed over the surgical field, and additional sterile (four-cornered) drapes overlay the sides of the transparent and large table drape and extend over the edges of the table and the rest of the animal.

The surgeon dons surgical mask and clean, disposable laboratory jacket (or, for nonhuman primate surgery, hair cover and sterile, disposable surgical gown) and sterile gloves after hand scrubbing.

b. Describe methods used to sterilize instruments and protective clothing, including a description of approved liquid sterilants and instrument exposure time(s) required for each, if applicable.

Surgical instruments and implanted materials are steam sterilized by autoclave or gas sterilized with ethylene oxide in [Redacted by agreement] or chemically sterilized in a chemical sterilant and rinsed with sterile water before use.

When used for sequential rodent surgeries, instruments may be disinfected, rinsed and instrument tips are heat sterilized in a hot bead sterilizer before and between rodent surgeries. The ACUC guideline, *Recommended Chemical Sterilants and Disinfectants*, advises investigators to use these chemical agents and manufacturer recommended contact times:

Recommended Sterilant	Brand name	Contact Time
Glutaraldehyde	Cidex® Activated Dialdehyde Solution, Cidex® Plus 28 Day Solution, Metricide® Activated Dialdehyde Solution, Metricide® 28 Long-Life Activated Dialdehyde Solution	10 hours
Chlorine dioxide	Clidox®, Alcide®, C-dox®	6 hours

c. Describe methods for instrument re-sterilization between serial surgeries.

Sterile instruments are required for all survival surgery. For "tips-only technique" in rodent surgery, instrument tips can be re-sterilized during and between serial surgeries

through heat (e.g. hot bead sterilizer) followed by adequate time or quenching in sterile water for the instrument to cool.

d. Indicate how effectiveness of sterilization is monitored.

The effectiveness of autoclave and ethylene oxide sterilization is confirmed by:
- Spore indicator and temperature sensitive tape (autoclave), and
- Spore indicator and gas sensitive paper indicator (Ethylene oxide).

Effectiveness of chemical sterilization is ensured by providing adequate contact times and use of freshly constituted solutions. Solutions are dated by date of constitution and or dispensing into an instrument container.

e. Describe surgical support functions provided by the program to investigators.

The animal program provides surgical support services (anesthesia, surgeon assistance, postoperative recovery and monitoring) to nonhuman primate surgeons.
The primary surgeon who performs chronic intravascular catheterization in Squirrel monkey has over thirty years of experience with implanting the external and internal jugular veins, femoral veins, and external iliac vein and is currently training another two surgeons.
Although not performed at NIDA yet, the primary surgeon who performs surgical placement of chambers for intracranial procedures and intravascular catheterization in Rhesus monkeys has twenty-five years of experience.

5. Intraoperative Monitoring [*Guide*, p. 119]
Describe monitoring and recording requirements for each species, including the type of record(s) maintained. Also note monitoring of anesthesia during non-survival procedures.

NHPs
The adequacy of anesthesia in nonhuman primates for surgical and research imaging procedures is evaluated by monitoring vital signs with electronic equipment and visual observation and evaluation of pain reflexes. Measurement of heart rate (obtained from electrocardiogram recordings as well as continuous pulse oximetry), respiratory rate, blood oxygen saturation, rectal temperature, end-tidal carbon dioxide and, when monitored, indirect blood pressure, is recorded at 10-15 minute intervals. The anesthetic record is added to the animal's individual medical record binder and or research data binder.

NHPs Non-survival - No non-survival surgical procedures are performed.

Rodents
Anesthetic monitoring in mice and rats is accomplished by observation of respiration and evaluation of pain reflexes, typically the rear toe-pinch reflex. Research personnel, certified as trained in the proper use of anesthetics, analgesics, and tranquilizers by the PI

(ASP Section N, item 4), monitors anesthesia of rodents during surgery. Rodent anesthesia records are limited and assimilated into the surgical record, e.g. date, anesthetic drug dose, surgery type and location, which are recorded within laboratory notebooks.

Rodents Non-survival

The adequacy of anesthesia for acute, non-survival rodent procedures involving surgery is monitored as described above for mice and rats. The adequacy of anesthesia for chronic, non-survival rodent procedures involving surgery, e.g. catheterization followed by fMRI imaging, is evaluated by monitoring vital signs, e.g. measurement of heart rate, respiratory rate, oxygen saturation, rectal temperature, with electronic equipment. Anesthesia recordkeeping practices are the same as for survival rodent surgery.

6. **Postoperative Care** [*Guide*, pp. 119-120]

Describe the postoperative care program, including who is responsible for overseeing and providing the care, types of records maintained (e.g., perioperative), where the records are maintained, etc.

Nonhuman Primates

Nonhuman primates are extubated and monitored in the surgery suite by the veterinary care staff until the animal is capable to maintain itself upright in its home cage.

- Before surgery, NHPs are food fasted overnight; upon recovery from anesthesia, primates are offered half of their daily food ration and returned to full ration on the next day.
- The veterinarian or animal health technician observe primates at least twice daily throughout the early (e.g., 3 days) postsurgical period for assessment of pain and the healing of the surgical site and then daily for the first week. The animal care staff also monitors postsurgical animals as part of their twice daily animal morbidity and mortality observations.
- For NHPs, a concise surgical record is created and included, along with the anesthesia record, as a hard copy attachment into the animal's individual medical record after surgery. Postoperative observations are recorded and added to the individual medical record, and filed in the room Redacted by agreement for use in assessment of surgical outcomes.

Mice and Rats

Rodents recover from surgery in a clean cage with soft, hardwood contact bedding, and are monitored until able to right themselves and remain in sternal recumbence.

- Designated research staff, typically the surgeon, provides primary postsurgical monitoring and care for at least 3 days but must continue to be observed throughout the early postsurgical period until the animal behaves normally and the incision is healing uneventfully. The animal care staff also monitors postsurgical rodents as part of their twice daily animal morbidity and mortality observations.
- Observed post-operative complications, including pain and distress, are treated, as per ASP, in consultation with the veterinarian.

- The surgeon or research technician records daily postsurgical rodent health observations and therapeutic treatments on a post-operative card which is maintained in the cage's card holder. The postsurgical care record is maintained in the animal's cage card holder for the acute period of postoperative observation, and investigators may later archive the record, typically within the animals' laboratory.

E. Pain and Distress [*Guide*, pp. 120-121]

1. Describe how and by whom pain and distress are assessed.

- The actual level of post-procedural pain and/or effectiveness of relief is assessed daily by animal and veterinary care staff during routine animal health observations.
- In addition to close observation of animals during or immediately after exposure to potentially nociogenic or anxiogenic experimental manipulation, review of short term data trends from operant behavioral testing, which can be a very sensitive indicator of the presence of even subtle pain and distress, can prompt researchers to request veterinary evaluation.
- The ACUC and PAV program also assess post-procedural pain in animals by observation of animals during their, respectively, semiannual inspection of the animal facilities and ASP compliance reviews.

2. Describe training programs for personnel responsible for monitoring animal well-being, including species-specific behavioral manifestations as indicators of pain and distress.

The ARAC Guideline, Guidelines for Pain and Distress in Laboratory Animals: Responsibilities, Recognition and Alleviation, and species-specific standard operating procedures contain provide guidance to staff to assess pain and distress through evaluation of changes in food or water intake, activity level, including species-specific behaviors, and surgical site complications that are suggestive of discomfort.

Research scientists are knowledgeable and PIs provide awareness training to their staff to regularly assess their animals' behavior and condition, through direct observation during daily handling and changes in testing behaviors, to suspect pain, distress, or illness which can confound their studies.

The Animal care staff are trained in recognition of animal pain and distress in context of their AALAS certification training program, veterinarian provided training in recognition of signs of illness, and corporate animal welfare training topics.

F. Anesthesia and Analgesia [*Guide*, pp. 121-123]

1. List the agents used for each species.
Note: If preferred, this information may be provided in Table or additional Appendix.

These pharmacologic agents have been approved in ASPs and SOPs:

Anesthetics	**Rats**	**Mice**	**NHPs**
Bupivacaine	X	X	X
Chloral hydrate	X	X	
CO_2 (terminal procedure only)	X		
Equithesin	X	X	
Isoflurane	X	X	X
Ketamine (+Xylazine)	X	X	X
Lidocaine (local)	X	X	X
Methohexital	X	X	X
Pentobarbital	X	X	
Telazol (+Xylazine)	X		
Urethane	X	X	
Analgesics			
Acetaminophen	X	X	X
Bupivacaine	X	X	X
Buprenorphine	X	X	X
Carprofen	X		X
Medetomidine	X (+Ketamine)		X (+Isoflurane)
Flunixin	X	X	X
Ibuprofen (Advil)	X	X	X
Ketoprofen	X	X	X
Lidocaine (Local)	X	X	X
Meloxicam	X	X	X
Xylocaine (Local)	X	X	
Xylazine(+Ketamine)	X	X	

2. Describe how the veterinarian provides guidance and advice to researchers concerning choice and use of anesthetics, analgesics or other pain moderating methods.

Veterinary guidance for selecting analgesic and anesthetic agents is provided both by consultation with the APD regarding specific application during ASP preparation, during subsequent ACUC review by fellow investigators, and during implementation of research experiments.

The APD updates the NIDA ACUC guidelines "Analgesia Guidelines" and "Anesthesia Guidelines for Rodents" to reflect contemporary, best analgesic practices to guide investigators during protocol development in determining the need for analgesia and choice of analgesic drugs for commonly performed surgeries. These guidelines will be available for the site visitors.

3. Describe the monitoring of the effectiveness of analgesics, including who does the monitoring. Include in the description any non-pharmacologic means used to diminish pain and distress.

Monitoring Analgesic Effectiveness

- The actual level of post-procedural pain and/or effectiveness of relief is assessed daily by animal and veterinary care staff during routine animal health observations.
- Designated research staff, typically the surgeon, provides primary postsurgical monitoring and care which includes administration of ASP prescribed analgesics and pain assessment.

Non-pharmacologic Relief

Non-pharmacologic means are employed to diminish fear and anxiety from procedures which can result in transient, mild discomfort, e.g. injection, as well as more pain, e.g. surgery.

- Many rodents are directly handled by research staff (gentled) on a daily basis, at times for weeks, before starting an experiment to reduce animal distress which can confound sensitive operant behavioral studies.
- Similarly, positive reinforcement training is used during training of all nonhuman primates to reduce the degree of potentially painful force needed for manual restraint by hand or collar-pole methods and to passive chair restraint before starting an experiment.
- Surgeons are trained to position animals, and, to avoid unnatural or unnecessary pressure or stress on body surfaces and joints for limb restraint, and to handle and manipulate tissues to minimize tissue trauma which can result in unnecessary post-procedural pain.
- Rodents are commonly housed individually after surgery in clean, dry bedded cages to prevent damage to implanted materials and to prevent animal injury from another potentially painful or distressed, postsurgical rodent.
- Supplemental environmental warmth and warmed, parenteral fluids may be provided perioperatively to reduce physiologic stress and potential enhancement of distress during recovery from anesthesia and surgery.

4. Describe how the veterinarian(s) and the IACUC/OB evaluate the proposed use of neuromuscular blocking agent to ensure the well-being of the animal.

Veterinary guidance for use of neuromuscular blocking agents is provided both by consultation with the APD during ASP preparation and further evaluated during subsequent ACUC review with special attention to adjunctive anesthesia use and pain monitoring.

- Neuromuscular blocking agents are not used in any currently active Animal Study Proposal.
- When proposed for use, the ACUC evaluates the scientific rationale, e.g. to prevent any stimulus-induced, return to consciousness during fMRI studies, and the

monitoring regimen, e.g. assessment of the animal's depth of anesthesia prior to administering the neuromuscular blocking agent and continual monitoring of heart rate, respiratory rate, oxygen saturation, rectal temperature, and tail cuff blood pressure to ensure a deep level of anesthesia.

5. Describe policies and practices for maintaining and ensuring function of equipment used for anesthesia.

The Animal Program sponsors and coordinates a biannual anesthetic vaporizer calibration certification, and the ACUC inspects vaporizers' date of inspection labels and adsorption (f/air) canisters monitoring activities during their semi-annual facilities' inspections.

The DOHS chemical hygienist monitors the effectiveness of devices for scavenging volatile anesthetics which include use of fume hoods and down-draft tables, local exhaust ventilation (snorkel) devices, and adsorption (f/air) canisters.

G. Euthanasia [*Guide*, pp. 123-124]

1. Describe approved methods of euthanasia, including humane slaughter (for additional guidance, see pertinent AAALAC Reference Resources). Include:

- consideration of species, age, condition (e.g., gestational period, or neonatal) and
- location(s) for the conduct of the procedure.

Note: If preferred, this information may be provided in Table or additional Appendix.

Considerations

The ACUC considers the species, age, and condition of animals by applying these guidelines for review and approval of euthanasia methods in ASPs and SOPs:

- AVMA Guidelines on Euthanasia
- NIH ARAC Guidelines
  - Guidelines for the Euthanasia of Rodent Fetuses and Neonates
  - Guidelines for Euthanasia of Rodents Using Carbon Dioxide

The ACUC may approve waivers from anesthesia use for physical methods based on scientific justification, provided by the PI in the ASP.

Approved Methods

A tabulated description of ACUC approved methods is:

Species	Methods
Mice	Cervical dislocation
	Anesthetic overdose of barbiturate, isoflurane, Equithesin, xylazine/ketamine followed by decapitation or thoracotomy with perfusion.
	Decapitation
	CO_2 overdose followed by cervical dislocation (adult mice), decapitation (neonatal mice) or thoracotomy.
	Fetuses – decapitation under residual CO2 or anesthesia of dam

	Exsanguination under anesthesia (perfusion fixation)
Rats	CO_2 overdose followed by cervical dislocation (neonatal rats), decapitation (<100 g) or thoracotomy.
	Cervical dislocation for rats < 100 g
	Decapitation
	Exsanguination under anesthesia (perfusion fixation)
	Fetuses – decapitation under residual CO2 or anesthesia of dam
	Anesthetic overdose of barbiturate, isoflurane, chloral hydrate, Equithesin, xylazine/ketamine followed by decapitation or thoracotomy with perfusion.
NHPs	After ketamine induction, barbiturate overdose which may be followed by thoracotomy with perfusion, as per ASP.

Locations

Euthanasia locations are visited and evaluated during ACUC semiannual review of the animal program. Euthanasia may be performed in the Redacted by agreement centralized animal facility necropsy rooms Redacted by agreement, which provide carbon dioxide tanks with pressure/flow regulators, timer, chamber lid, and dedicated guillotines and necropsy scissors, and down-draft stainless-steel necropsy tables with perfusate collection containers, or in approved investigator laboratories.

Redacted by agreement are the only locations for CO2 euthanasia of rodents, commonly performed by the animal care staff.

- Euthanex® euthanasia equipment with wall mounted CO2 cylinders are provided.
- CO2 euthanasia instructions are visibly posted to supplement user training.

Research technicians also perform ASP specified euthanasia of rodents followed by perfusion or tissue harvest in shared rooms Redacted by agreement and Redacted by agreement Appropriate portable monitoring equipment from the survival surgery suite is redeployed to monitor and confirm death for NHP perfusions in Redacted by agreement Additionally, Redacted by animal laboratory room Redacted by agreement contains downdraft stainless steel necropsy tables for rodent perfusion.

Physical and chemical methods of rodent euthanasia are performed in these ASP identified animal laboratory locations:

Redacted by agreement

Redacted by agreement	

2. Describe policies and practices for maintaining and ensuring function of equipment used for euthanasia.

The ACUC checks euthanasia equipment during semiannual inspection of animal areas and recalls investigators' guillotines every two years to assess sharpness and, when needed, to coordinate animal program-sponsored sharpening by a commercial vendor.

The animal care staff is trained to review the pressure gauge on the carbon dioxide tanks to change and or reorder tanks before exhaustion. Posted, illustrated instructions regarding operation of pre-set flowmeters and dual-event timer are used for training animal care and research staff to ensure consistent and adequate duration of exposure to slow and rapid gas flows.

3. Describe the methods used to confirm death of an animal.

Death of an animal is confirmed by absence of palpable heart beat, prolonged cessation of breathing, and absence of response to pain reflexes.

A redundant euthanasia method is practiced following CO2 euthanasia: cervical dislocation is performed on adult mice and cervical decapitation on neonatal mice (scissors) and rats (guillotine).

IV. Physical Plant [*Guide*, pp. 133-155]

A. Facilities Overview

Provide a brief introduction to the animal housing and use facilities. Note that this overview should augment the information provided in **Appendix 2** (Summary of Animal Housing and Support Sites), which includes area, average daily census, and person responsible for each site. Please use consistent terminology for the buildings/areas/sites described in the Location section of the Appendix. Please do not repeat information, but supplement the descriptions provided elsewhere to assist the reviewers understanding of the interaction between facilities, special housing locations, and separate procedural areas.

NIDA occupies portions of the Redacted by agreement Redacted by agreement, on the Johns Hopkins Bayview campus, Baltimore, MD.

Redacted by agreement Redacted by agreement contains the NIDA and National Institute on Aging (NIA) intramural research programs who independently operate centralized animal facilities on separate floors. NIDA and NIA share a bulk cage wash chemical tank area Redacted by agreement a loading dock with an indoor MPW cold storage area Redacted by agreement and an outdoor compressing waste dumpster, and an outdoor vacuum bedding waste dumpster. The centralized NIDA animal facility is located on the Redacted by agreement Redacted by agreement dedicated NIDA "clean" and shared NIA "dirty" animal loading docks.

Redacted by agreement NIDA is the only entity which operates animal facilities in the Redacted by agreement Redacted by agreement and ACUC approved mouse satellite housing areas Redacted by agreement Redacted by agreement within a research laboratory suite Redacted by agreement NIDA and other tenants share a freight elevator, used for animal transport to laboratories, and a common loading dock with a compressing waste dumpster. A NIDA guideline describes practices for operation of the satellite housing area outside of the centrally-managed Redacted by agreement animal facility.

NIDA guidelines also describe practices for housing rodents for more than 24 hours within Redacted by agreement vivarium study areas. The Redacted by agreement animal facility contains separate suites of multiple-user rooms, each under oversight of a lead laboratory, that are dedicated for ASBL-2 containment, radioisotope containment for conduct of PET studies, and a fMRI animal study area.

B. Centralized (Centrally-Managed) Animal Facility(ies)

In this section, describe each centralized or centrally-managed animal housing and use facility. Include in **Appendix 3** the floor plans of each on 8.5" x 11" or A4 paper. Ensure that the drawings are legible and the use of each room is indicated (animal housing, procedure room, clean cage storage, hazardous waste storage, etc.). Note that a separate section for describing "satellite housing areas" is included below.

Separately describe **each** Location or Animal Facility, addressing each of the features outlined below (1-8). A complete description of each must be provided; however, common features among locations or facilities may be indicated as such and do not need to be repeated.

1. General arrangement of the animal facilities (conventional, clean/dirty corridor, etc.).
2. Physical relationship of the animal facilities to the research laboratories where animals may be used.
3. Types of available animal housing spaces used, such as conventional, barrier, isolation/quarantine, hazard containment (infectious, radioactive, chemical), "animal cubicles" or facilities specifically designed for housing certain species such as ponds, pastures, feedlots, etc.

4. Finishes used throughout the animal facility for floors, walls, ceilings, doors, alleyways, gates, etc. (note any areas that are not easily sanitized and describe how these are maintained).
5. Engineering features (design, layout, special HVAC systems, noting exhaust air treatment, if applicable) used in hazardous agent containment.
6. Security features, such as control of entry, perimeter fences, gates, entryways, cameras, guards; identify and describe exceptions for individual facilities or areas incorporating fewer or additional security features than the general features described.
7. Consideration for facilities with exterior windows, if applicable, including management of environmental conditions (i.e., temperature and photoperiod control) and potential security risks.
8. Storage areas for flammable or hazardous agents and materials (e.g., disinfectants, cage-washing chemicals, pesticides, fuel).

General Arrangement

The Redacted by agreement animal facilities are managed as a conventional, SPF facility with a single corridor system and bi-directional traffic pattern to animal housing, procedure, and support rooms.

Physical Relationship between Animal Facilities and Research Laboratories

Redacted by agreement	Redacted by agreement
With a few exceptions on the upper floors of the Redacted by all animal research laboratories, including laboratories in which rodents are housed to satisfy behavioral testing or radioisotope or ABSL-2 containment requirements, are contiguous to the centralized housing areas and on the Redacted by agreement	NIDA animal research laboratories are located contiguous to the Redacted by agreement animal facility and clustered on the Redacted by agreement Redacted by agreement

Types of Animal Housing Spaces Used

Redacted by agreement	Redacted by agreement
In additional to conventional housing of mice, rats, Squirrel monkeys and Rhesus monkeys, areas are designated for mouse and rat housing for quarantine holding, containment holding during use of radiologicals (PET suite) and during exposure to agents used with ASBL-2 and ASBL-3 practices (ASBL suite), and greater than 24 hours holding for behavioral testing in animal laboratories within the animal facility. PET suite and ASBL suite is described separately, below, in Section E.1. Other Animal Use Facilities.	All housing within the centrally managed animal facility is conventional housing of rats and mice. A satellite facility, described separately, below, in Section C. Satellite Animal Housing Facilities, is located within Redacted by agreement and conventionally houses mice.

Rodent quarantine in performed in Redacted by agreement Redacted by agreement a dedicated room with a negative differential pressure to adjacent areas. The quarantine room contains Redacted by agreement Redacted by agreement (B&H, Inc. Isolation and Containment Cubical) for housing. The contiguous corridor contains two-sided autoclaves for material pass-through to dirty-side cage wash and an anteroom for personnel and rolling stock (racks, carts) to exit to the dirty side Cage Wash.	

Finishes

Redacted by agreement	Redacted by agreement
Floors • Troweled, seamless epoxy resin coved to a height of 6 inches at the wall/floor junction in vivarium housing rooms, animal laboratories, non-rodent surgery suite, support rooms, and personnel rooms. • Methylmethacrylate (MMA) floors in the high traffic cagewash rooms • Seamless sheet vinyl in animal testing laboratories outside of the vivarium perimeter.	• Troweled, seamless epoxy resin coved to a height of 6 inches at the wall/floor junction in vivarium housing rooms and animal laboratories, and support rooms. • Methylmethacrylate (MMA) floors in the high traffic cagewash rooms • Seamless sheet vinyl in vivarium personnel rooms and animal testing laboratories outside of the vivarium perimeter.
Walls • Epoxy paint finish over filled masonry block and over gypsum panels and metal frame walls in corridors, animal laboratories, and centralized housing areas. • Wall mounted aluminum guardrails and stainless-steel corner guards protect, respectively, animal facility corridor walls, corners and doorframes.	• Epoxy paint finish over masonry block in Redacted by agreement vivarium animal laboratories and over gypsum panels and metal frame walls in animal laboratories outside of the vivarium. • Wall mounted aluminum guardrails and stainless-steel corner guards protect, respectively, animal facility corridor walls, corners and doorframes. • Floor to ceiling fiberglass reinforced plastic (FRP) panels over filled masonry block and over gypsum panels and metal frame walls in centralized animal housing, cage wash, and support rooms.
Ceilings • Suspended, scrubable fiberglass reinforced plastic (FRP) tiles are used in animal housing, support, cage wash, and testing areas in the animal facility. • Epoxy paint finish over gypsum panels in animal housing, testing, and support areas.	• Suspended, scrubable 2'x4' vinyl-faced ceiling panels animal laboratories outside of the vivarium. • Water resistant fluorescent lighting banks are recessed.

• Water resistant fluorescent lighting banks are recessed.	
Doors • Epoxy paint finished galvanized steel doors or fiberglass doors in a heavy gauge steel frame with hinged observation windows are used in all vivarium rooms; behavioral testing room doors are galvanized sound control doors with galvanized frame. • Sealed wooden doors and galvanized steel frames are used for animal laboratories outside of the vivarium.	• Fiberglass doors with hinged observation windows in a heavy gauge stainless steel frame are used for housing rooms. • Sealed wooden doors and galvanized steel frames are used for animal laboratories, outside of the vivarium.

Behavioral laboratories are typically outfitted wall mounted Corian® shelves or mobile, castered racks containing open field chambers or sound attenuating chambers with ancillary computerized data collection equipment; a few behavioral testing laboratories contain typical, general laboratory closed, wall and base cabinets with countertops for microdialysis and open-field chambers.

The single satellite housing application Redacted by agreement houses mice in ventilated cage racks (Innovive®) for testing within sound attenuating chambers with ancillary computerized data collection equipment on mobile, castered racks in the satellite room. Animal room temperature and photoperiod is controlled and monitored remotely by the animal care staff through the automated building system.

Engineering Features Used in Hazardous Agent Containment

The building automation system continually monitors supply and exhaust air flows and temperatures of room exhaust air. Room level control of supply air flow is programmed in the Redacte to trail exhaust air flow to maintain critical negative differential air pressures, e.g. ABSL-2, PET, NHP housing and testing, and quarantine suites, between room and corridor, i.e. negative pressures are preserved as ventilation rates decrease towards a static, neutral pressure state.

Ceiling mounted and table top, local exhaust ventilation (LEV) devices are used in many Redacted by agreement rodent surgery areas for scavenging waste anesthetic gases and dental cement organic vapors to room HVAC exhaust ducts. A dedicated vacuum system exhausts waste anesthetic gas in the Redacted by agreement

The Redacted by agreement contains an oxygen sensor, maintained by the Building managers, which alarms when oxygen levels fall below normal limits (18%). Helium cryogen is contained within the fMRI magnet in a primary container whose pressure relief valve

releases gas into a secondary container from which gas escapes through an 18" diameter, heavy gauge stainless steel quench pipe directly to the outdoors. The fMRI rooms maintain a negative differential pressure to the control room and again to the personnel corridor.

Security Features

Redacted by agreement

Redacted by agreement

Storage Areas for Flammable or Hazardous Materials

Flammable or hazardous agents are stored in Safety approved, storage cabinets in room Redacted by agreement (Ethylene oxide canisters, isopropyl alcohol) and within research laboratories within the vivarium.

- Cage wash chemicals are dispensed from double-walled bulk tanks in a shared Redacted by agreement Redacted by agreement
- Additional sanitizing chemicals are stored within Redacted by agreement segregated within Redacted by agreement and dispersed amon Redacted by agreement and Redacted by agreement Redacted by agreement

There are no exterior windows in animal rooms. Door mounted windows for housing and testing rooms have covers.

C. Satellite Animal Housing Facilities

In addition to the Appendices summarizing Heating, Ventilation, and Air-Conditioning (**Appendix 11**) and Lighting Systems (**Appendix 16**), summarize animal housing areas that are not centrally-managed or maintained in (**Appendix 17**), "Satellite Animal Housing Areas."

1. Describe the criteria used to determine/define a "Satellite Animal Housing Area," which may include remote housing facilities or laboratories temporarily or consistently housing animals.

The ACUC defines satellite animal housing areas as locations outside of the centrally managed animal care and use facility which temporarily or consistently house animals for

more than twenty-four hours. There is one location where animals are housed in a satellite arrangement, Redacted by agreement

In the NIDA program, rats and mice may be held for more than twenty-four hours in animal laboratories that are located within the perimeter of the centrally managed vivarium. These laboratories were designed into the original vivarium construction and are served by the same HVAC system. Research staff provide daily animal care following ASP and NIDA SOP and ACUC guidelines.

2. Describe the process used by the IACUC/OB to authorize, provide oversight of, and ensure compliance with *Guide* standards for the housing of animals outside of centrally-maintained facilities. Include a description of Attending Veterinarian access and physical security.

- The ACUC reviews and approves justified need for satellite housing occurs at the time of ASP review and customized descriptions of standard operating procedures for husbandry, sanitation, and documentation prior to animal occupation.
- The Attending Veterinarian (APD) determines the space's suitability regarding HVAC, security, environmental (temperature, humidity, photoperiod) control and monitoring, and pest management and communicates required renovations through NIDA administrators to the Building Managers.
- The APD, facility veterinarian, and contract husbandry staff have authorized card key access. Initially, satellite housing is reviewed by frequent visits by the veterinary and husbandry staff, and once housing practices are well established, is reviewed by ACUC during semi-annual facility inspections.
- The satellite facility security is described in Section B. Centralized Animal Facilities, two pages earlier, in the description of the Redacted by agreement and "Animal Laboratory" subsections.

D. Emergency Power and Life Support Systems

Note: Complete a Heating, Ventilation, and Air-Conditioning (HVAC) Summary (**Appendix 11**) and Lighting Summary (**Appendix 16**) for each Location described in the Summary of Animal Housing and Support Sites (**Appendix 2**).

1. Power [*Guide*, p. 141]

For each Location, Centralized Animal Facility, and Satellite Housing Facility, provide a brief description of the following:

- Availability of emergency power and if so, what electrical services and equipment are maintained in the event the primary power source fails.
- History of power failures, noting frequency, duration, and, if emergency power was not available, steps taken to ensure the comfort and well-being of the animals present and the temperature extremes reached in animal rooms during the failure.

Redacted by agreement	Redacted by agreement
Emergency Power	

• On-site electrical generators provide power upon loss of electric utility feed to support the BAS control systems, air handlers, egress corridor lighting, animal cubicles in quarantine and ABSL-2 areas, and red colored, emergency powered receptacles serving ventilated rack HEPA supply blowers in housing rooms and animal laboratory equipment. • Uninterruptable power supply units protect BAS control systems and the housing rooms' Enviro-Gard™ MultiPlex™ supply air blowers against surge related interruption during power change overs. • Emergency power is not provided to HVAC chillers and animal housing and testing room lighting.	• On-site electrical generators provide power upon loss of electric utility feed to support the BAS control systems, air handlers, chillers, steam boilers, corridor and room lighting and receptacles serving ventilated rack blowers. • Uninterruptable power supply units protect BAS control systems and the housing rooms' Enviro-Gard™ MultiPlex™ supply air blowers against surge related interruption during power change overs.
Power Failures NIDA has occupied the [Redacted by agreement] [Redacted by agreement] since 2008. Failures to the [Redacted] power occurred: • 2008: June (~ 1 hour duration) and July (two ~ 1 hour duration incidents) • 2011: October ~ 6 hour duration • 2018: December ~ 9 hour duration The [Redacted by agreement] has dual feed lines from a single substation. Brief power failures of seconds to minutes duration are readily compensated by the building's generators without impact on animal well-being and comfort.	NIDA has occupied the [Redacted by agreement] since 1993. Failures to the [Redacted] power occurred: • 2003: ~ 5 hour duration • 2012: June, ~ 2 hours (emergency capacity hadn't yet extended to include chillers) Brief power failures of seconds to rarely minutes duration to the single [Redacted by agreement] power feed is common. Subsequent to 2003, emergency power generation capacity was incrementally increased to the current 2.1 megawatts, which exceeds the utility's feed and supports the entire building including the two satellite housing areas.

2. **Other System Malfunctions.** If not previously reported, describe animal losses or health problems resulting from power, HVAC, or other life support system (e.g., individually ventilated cages) failures, and mechanisms for reporting such incidences. AAALAC International Rules of Accreditation (Section 2.f).

Planned utility system shutdowns, e.g. for repair, renovation, and maintenance of HVAC, electrical power, water systems are scheduled, whenever possible, and coordinated with

vivarium staff. There have been no animal losses or health problems resulting from power, HVAC, or other life support system failure.

The mechanism for reporting animal losses or health problems resulting from power, HVAC, or other life support system failures is identical for all animal welfare concerns, as described in Section I.A.1.2.c

E. Other Facilities [*Guide*, pp. 144, 150]

1. Other Animal Use Facilities [*Guide*, pp. 146-150]

Describe other facilities such as imaging, irradiation, and core/shared behavioral laboratories or rooms. Include a description of decontamination and methods for preventing cross-contamination in multi-species facilities.

Magnetic Resonance Imaging

The [Redacted by] animal facility contains a restricted access, functional magnetic resonance imaging (fMRI) suite for imaging rats, mice, and nonhuman primates. The suite contains a central 9.4T Brucker MRI [Redacted by agreement] with access only through the adjacent control [Redacted by agreement]. The MRI room is sheathed with radiofrequency and magnetic shielding. The control room is contiguous with a rodent surgery/animal preparation area [Redacted by agreement] and an adjacent rat behavioral testing [Redacted by agreement] and MRI equipment room [Redacted by agreement] surrounding [Redacted by agreement] The MRI suite contains equipment to provide isoflurane anesthesia, to scavenge waste anesthetic gases, to maintain animal warmth, and to ventilate and monitor anesthetized animals, oxygen level alarms, and a quench pipe to passively exhaust cryogen gas which escaped the MRI magnet core. The BAS controlled, supply and exhaust air flows can be increased during cryogen filling.

The MRI facility is managed by the Neuroimaging branch scientists and follows its own laboratory and ASP practices for sanitation of MRI equipment and are also designed to also prevent cross-contamination between species. Dedicated anesthesia, fMRI coils, animal bed, and other related equipment is used only for NHPs.

PET Imaging Suite

The [Redacted] animal facility contains a restricted access, positron-emission tomography (PET) suite for imaging rats, mice, and nonhuman primates. The PET suite contains separate areas for microPET imaging, rodent surgery, animal housing, and behavioral testing. The suite perimeter is sheathed within lead cladding.

The PET laboratory suite is managed by Biobehavioral Imaging and Molecular Neuropsychopharmacology unit scientists which [Redacted by agreement] [Redacted by agreement] ensures personnel training, monitors suite usage, and coordinates closely with the Safety, DRS, and the animal care program staff. Nonhuman primates are imaged separately from rodents, and the animal housing area is sanitized before introducing and after removal of NHPs. Dedicated anesthesia and other related equipment

is used only for NHPs. Scientists follows the ASP described practices for sanitation of microPET imaging and NIDA SOPs for sanitation of behavioral testing equipment.

BSL2/3 Containment Suite

The Redacted by animal facility contains a restricted access, BSL-2/3 containment suite for using agents with ABSL-2 and ABSL-3 practices. The suite consists of:

- Redacted by agreement with separate staging areas to dispense PPE, to store disposable mouse and clean rat caging, which are etched with "BSL2," to dispose of used PPE, stage used cages for further decontamination/cleaning, and a separate, designated "clean" cart for corridor transport between housing and ABSL-2 area and a "dirty/ABSL-2" cart for corridor transport between ABSL-2 and decontamination (autoclave, ethylene oxide) and dirty cage wash areas.
- Procedure room Redacted by agreement with laboratory assigned, lockable storage cabinets, flammable cabinet for alcohol storage, decontamination solutions, anesthetic vaporizer with scavenging system, postsurgery recovery chamber, HEPA vacuum cleaner, fur clippers, warm-water heating blanket, instrument cleaning, table top autoclave, etc to support rodent surgeries.
- A multifunction room Redacted by agreement containing a small sink and Redacted by agreement (B&H, Inc. Isolation and Containment Cubical), each dedicated to a separate function: 1) cage changing/material manipulation/injections within a biological safety cabinet, 2) mouse housing, 3) rat housing, and 4) to replace function of another cubicle in case of failure.

The Director of the Optogenetic and Transgenic Animal Core oversees implementation of the Core's standard operating procedures for using agents (e.g. lentivirus, AAV, BrdU, diphtheria toxin) within the animal facility's ABSL-2 suite. The Director verifies personnel training, reviews Recombinant DNA Registration for BSL2/3 Activities documentation, coordinates surgery schedules, and ensures the users provide daily husbandry for the duration of housing within the ABSL-2 suite. Specific methods for decontamination of equipment varies with the agent and are designed to also prevent cross-contamination between rats and mice within the ABSL-2 suite and between rodents within the suite and general experimental housing. Methods are described within the core's SOP and individual investigators' ASP.

Animal Program Shared Spaces

The centralized animal program provides a room in each animal facility for researchers to perform euthanasia, including terminal animal perfusion, and a room in the Redacted by agreement for rodent surgery. Sanitation practices follow NIDA SOPs for animal laboratory areas. The necropsy (euthanasia) rooms contain downdraft workstations for performance of CO2 euthanasia, paraformaldehyde perfusions, and tissue harvesting, and freezers to accumulate non-hazardous carcasses for entry into the MPW stream. Cleaning supplies are provided for investigators for postprocedural cleaning of workstation surfaces between users, and the animal care housekeeping staff provides additional sanitation support to the workstation and room. The electronic room reservation system is used to exclude other users and the door is posted to preclude entry during the infrequent occurrences of NHP perfusion.

2. Other Animal Program Support Facilities

Describe other facilities providing animal care and use support, such as feedmills, diagnostic laboratories, abattoirs, etc.

There are no animal use facilities specialized for NIDA use. The NIH's AAALAC accredited program provides facilities to support animal care and use for all institutes including NIDA for NHP quarantine at the NIH Animal Center (Poolesville, MD) and VRC Diagnostic laboratory and Pharmacy (Bethesda, MD)

According to the privacy principles on the protection of natural persons with regard to the processing of personal data and on the free movement of such data, we wish to advise you that the personal data in the Program Description will become part a permanent file owned by AAALAC International, and that can be shared with AAALAC International offices and representatives in order to perform an evaluation of the institution´s animal care and use program and provide accreditation services. The institution has the option of exercising rights of data access, rectification, cancellation, and opposition at: accredit@aaalac.org

Appendix 1: Glossary of Abbreviations and Acronyms

Please provide a Table defining abbreviations and acronyms used in this Program Description.

Abbreviation/Acronym	Definition
APD	Animal Program Director
ARAC	Animal Research Advisory Committee
ASP	Animal Study Proposal
AV	Attending Veterinarian
B virus	Cercopithecine herpesvirus 1
BNRB	Behavioral Neuroscience Research Branch
Redacted by agreement	Redacted by agreement
CNRB	Cellular Neurobiology Research Branch
DHHS	Department of Health and Human Services
DOHS	Division of Health and Safety
DVR	(NIH, Office of Research Services) Division of Veterinary Resources
fMRI	Functional Magnetic Resonance Imaging
IBC	Institutional Biosafety Committee
IO	Institutional Official
IRP	Intramural Research Program
LAAPP	Laboratory Animal Allergy Protection Program
MRI	Magnetic Resonance Imaging
MTMD	Molecular Targets and Medication Discovery Branch
NHP(s)	Nonhuman primate(s)
NIDA	National Institute on Drug Abuse
NIH	National Institutes of Health
NRB	Neuroimaging Research Branch
OHSC	Occupational Health and Safety Compliance branch

Appendix 1: Glossary of Abbreviations and Acronyms

Abbreviation/Acronym	Definition
OLAW	Office of Laboratory Animal Welfare
OMS	Occupational Medical Services
ORF	Office of Research Facilities
ORS	Office of Research Services
OSD	Office of the Scientific Director
OTTC	Optogenetics and Transgenic Technology Core
PAV	Protocol Adherence Verification program
PCR	polymerase chain reaction
PHS	Public Health Service
PI	Principal Investigator
PM	(NIH) Policy Manual

Appendix 2: Summary of Animal Housing and Support Sites

Redacted by agreement

Appendix 2: Summary of Animal Housing and Support Sites

Redacted by agreement

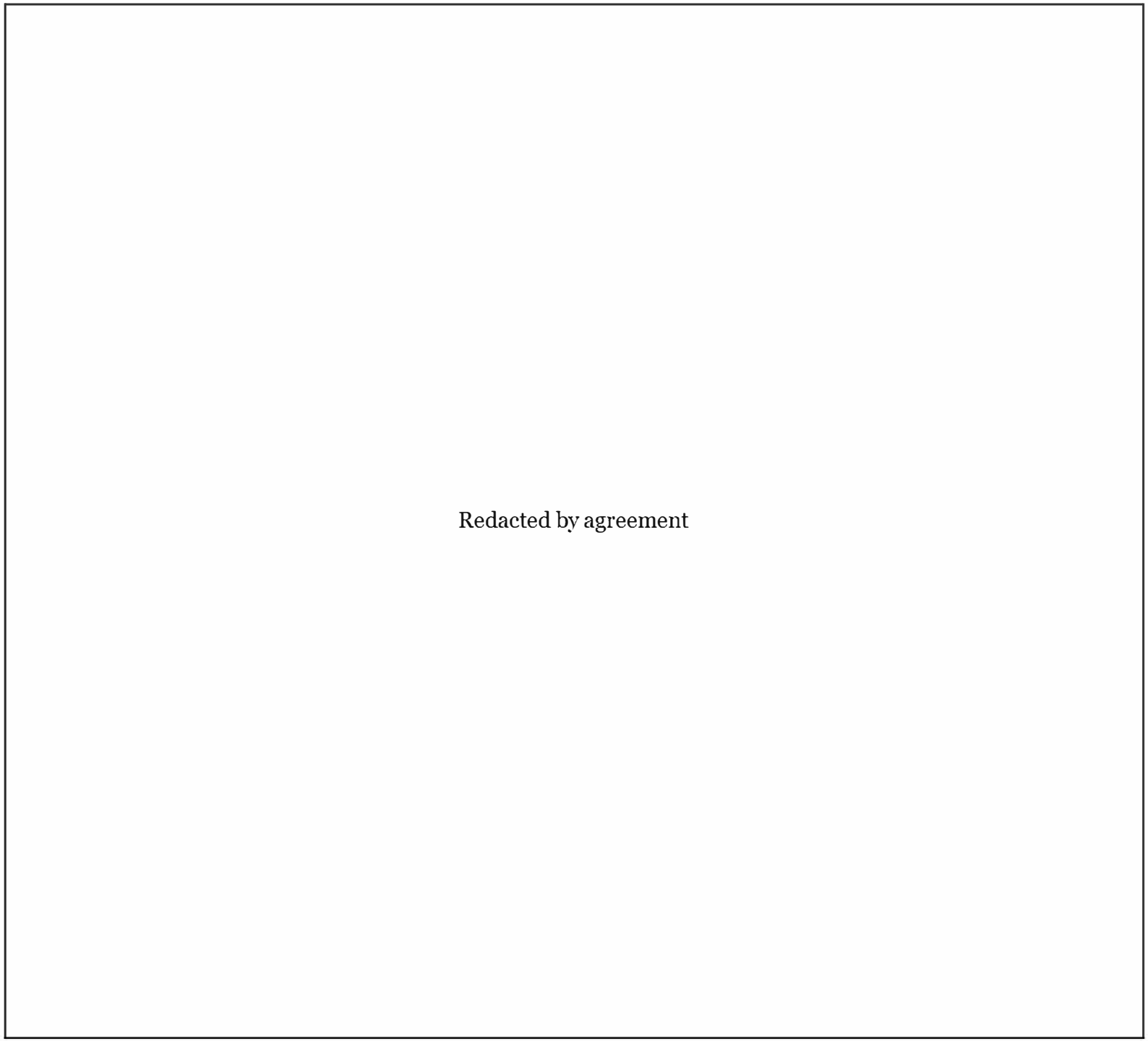
Appendix 3: Line Drawings

Redacted by agreement

Appendix 3: Line Drawings

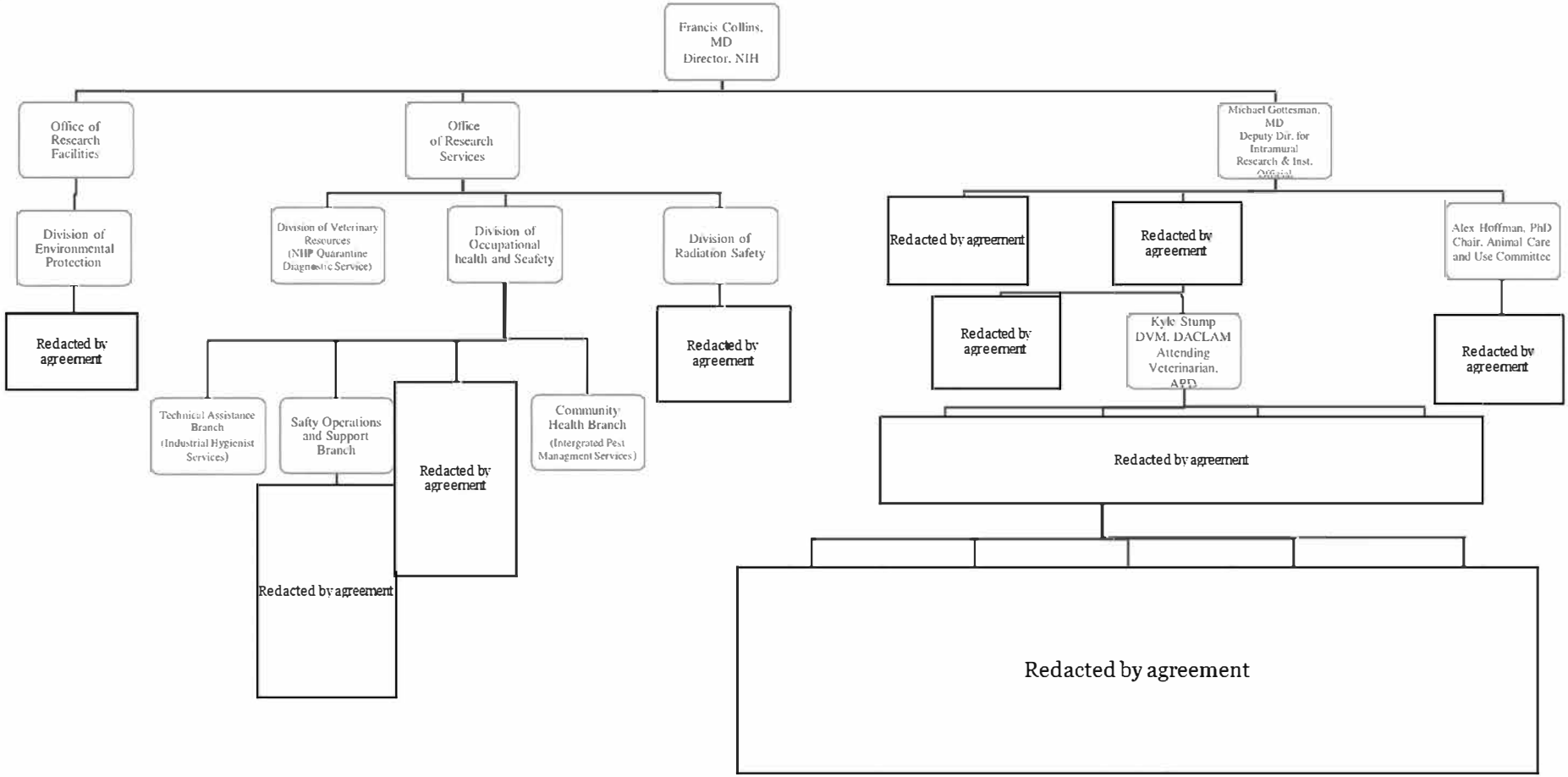
Redacted by agreement

Appendix 3: Line Drawings



Appendix 4: Organizational Chart(s)

Provide an accurate, current, and detailed organization chart or charts that detail the lines of authority from the Institutional Official to the Attending Veterinarian, the IACUC/OB, and personnel providing animal care. If applicable, include personnel responsible for managing satellite housing areas/locations and depict the reporting relationship between the Attending Veterinarian and other(s) having a direct role in providing veterinary care.



Appendix 5: Animal Usage

In order to assist the site visitors in their evaluation of the animal care and use program, please provide the information requested below. Information should be provided for all animals approved for use in research, teaching or testing, including those which may be used or housed in laboratories outside the animal care facility. Of particular interest is information on those animals which are used in research projects involving recovery surgical procedures, behavioral or other testing requiring chairing or other forms of restraint, or exposure to potentially hazardous materials. An alternate format is acceptable as long as the information requested is provided.

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
Effects of alpha-synuclein on toxicity in midbrain dopaminergic neurons	Redacted by agreement		Mice Rats	2320 3230	C, D	✓				✓	
Modulatory roles of neurotransmitter systems in the effects of addictive drugs in rodents			Rats	286	C, D	✓	✓	✓		✓	
Neural mechanisms underlying the incubation of methamphetamine, cocaine and heroin craving after voluntary abstinence			Mice Rats	40 1080	D	✓	✓	✓		✓	
Transcranial magnetic stimulation in squirrel monkeys			Squirrel Monkey	12	C					✓	

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
Role of prefrontal cortical neuronal ensembles in encoding extinction memory for cocaine self administration	Redacted by agreement		Rats	800	D	✓		✓			✓
In vivo calcium imaging in freely behaving rodents using a miniature epifluorescence microscope			Rats	2184	D	✓	✓	✓		✓	
Sensitized reaction to stressful events and its neural mechanisms in cocaine-experienced rats			Rats	1320	C, D	✓	✓			✓	
Determinants of nicotine and THC self-administration in squirrel monkeys			Squirrel Monkey	72	C, D	✓	✓			✓	
Rodent in vivo and ex vivo models to study receptor heteromers as targets for drug			Mice Rats		C, D	✓	✓			✓	

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
development in neuropsychiatric disorders				1960 944							
Effects of abused drugs on cardiovascular function in rats	Redacted by agreement		Rats	21	D	✓				✓	
The neuroprotective and neurorestorative effects of trophic factors in MPTP mouse model of Parkinson's disease\			Mice	1020	D	✓				✓	
Electrophysiological characterization of serotonin regulation of orbitofrontal cortical neurons in rodent brain slices			Rats	802	C, D	✓				✓	
Studies on SIGMA-1 receptors in primary cultures and brain slices			Mice Rats	268 320	C, D	✓					
Functional study of the VTA afferents			Mice		D	✓		✓			

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
from the parabrachial nucleus (PBN) or the pedunculopontine tegmental nucleus (PPTg)				1648			✓			✓	
Viral-mediated investigations of reward and aversion in mice and rats	Redacted by agreement		Mice Rats	8210 1720	D	✓	✓	✓		✓	
Identification of afferents to different cell types of the brain reward system			Mice Rats	2320 3230	C, D	✓	✓			✓	
Development of a model of operant drug vapor self-administration in rodents			Mice Rats	120 768	C					✓	
Effects of anti-abuse candidate drugs on the reinforcing effects of cocaine			Rats	270	D	✓	✓	✓			
Role of the microRNA system on dopaminergic and			Mice	980	C, D	✓					

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
behavioral effects of drugs of abuse											
Transcriptional profile of brain reward circuits following methamphetamine self-administration	Redacted by agreement		Rats	4110	D	✓	✓	✓		✓	
Modulation of the behavioral and neurochemical effects of psychostimulants by different brain receptor systems			Mice Rats	5000 8680	C, D	✓					
Neurobiology of newly-emerging stimulant drugs of abuse			Rats	2406	C, D	✓					
Changes to network connectivity in response to acute nicotine in nicotine-naïve and nicotine experienced rats.			Rats	48	D	✓				✓	
A pilot functional MRI study in an animal			Rats	58	C						

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
model of alcohol-schizophrenia comorbidity											
Development of autoradiography assay for imaging DREADDs in vitro in mice and rats	Redacted by agreement		Mice Rats	1311 655	C, D	✓				✓	
The effects of zinc availability on cocaine reward			Mice	2195	C, D	✓		✓		✓	
Monitoring methamphetamine-induced endoplasmic reticulum stress using cerebral spinal fluid (CSF)			Mice Rats	21 540	D	✓	✓			✓	
In vivo imaging of neural cluster activity for opioid reward			Mice	320	D	✓	✓			✓	✓
In vivo imaging of neuron activity accompanying motor and cognitive abnormality in			Mice	1232	C, D	✓	✓	✓		✓	✓

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
Parkinson's disease model mice											
Characterization of novel microglia-specific expression of cre-recombinase	Redacted by agreement		Rats	4890	C, D	✓				✓	✓
Using focused ultrasound for disruption of blood brain barrier: developing minimally invasive gene delivery to the rodent brain			Rats	608	D	✓				✓	✓
Neural circuits of motivation and reward in mice and rats			Mice Rats	6460 160	C, D	✓	✓	✓		✓	✓
Functional alterations in neuronal ensembles during food self-administration learning			Rats	552	C			✓		✓	
In vivo imaging of neuronal circuit activity for			Mice	1200	D	✓	✓	✓		✓	✓

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
psychostimulant reward											
In vivo imaging of dorsal striatal circuits during the acquisition and consolidation of a motor learning skill	Redacted by agreement		Mice	544	D	✓	✓			✓	✓
In vivo imaging and electrophysiolgical studies of hypothalamic neuronal circuits controlling motivated behaviors			Rhesus	4	D	✓	✓	✓		✓	
In vivo ensemble labeling in freely behaving rodents using a genetically encoded photoconvertible calcium integrator			Rats	3278	D	✓				✓	✓
Primate model for development of PET radioligands for DREADDS			Squirrel monkey	4	D	✓	✓			✓	

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
Effects of psychostimulants on brain temperature homeostasis	Redacted by agreement		Rats	230	D,E	✓				✓	
Neurochemical correlates of addictive and anti-addictive drug actions using microdialysis and optogenetics			Rats	3505	C, D	✓				✓	
Behavioral and Neurochemical Effects of Addictive and Anti-addictive Drugs in Transgenic Mice			Mice	17680	C, D	✓		✓		✓	
Prestnaptic and postsynaptic connectivity of midbrain neurons			Rats	1915	C, D	✓				✓	
Hypothalamic neuronal circuits controlling motivated behaviors			Mice	3579	D	✓		✓		✓	✓
Modifying microglia in the rodent brain to			Rats	2144	D, E	✓		✓		✓	

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
reduce neuroinflammation											
Role of Lateral Habenula in Cocaine Self-administration Behavior	Redacted by agreement		Rats	936	D	✓	✓	✓		✓	
Connectivity of hypothalamic neuronal circuits controlling motivated behaviors			Mice	3018	D	✓		✓		✓	
In vivo imaging and electrophysiolgical studies of hypothalamic neuronal circuits controlling motivated behaviors			Mice	4920	D	✓		✓		✓	
Role of VTA endocannabinoids on cocaine self administration in mice			Mice	504	D	✓	✓			✓	
In vivo calcium brain imaging during			Rats	340	D	✓	✓	✓	Acc, 90 min	✓	

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
predictive learning in rats											
Neurotoxic effects of methamphetamine: the role of DNA methylation and potassium channels	Redacted by agreement		Rats	2740	C, D	✓				✓	✓
Psychostimulants-induced neuroplasticity			Mice	2640	C					✓	
Role of Sigma-1 receptor on neuropathic pain			Mice Rats	376 472	E	✓				✓	
Holding protocol for transgenic rats			Mice	2880	C, D					✓	
Viral-mediated investigations of neuronal circuits of pain modulation and opiate reward in mice			Mice	3860	C, D, E	✓	✓	✓		✓	
Evaluation of the involvement of glycine receptors on opioid			Mice	1040	C, D, E	✓	✓				

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
self-administration in male and female mice											
Oral Opioid Self-Administration	Redacted by agreement		Rats	504	C					✓	
Microdialysis screening of candidate drugs for cocaine antagonist activity			Mice Rats	9300 19170	C, D	✓				✓	
Study of receptor heteromers in ex vivo mouse models for drug development in neuropsychiatric disorders			Mice	630	C, D	✓				✓	
Effects of potential medical treatments for cocaine use disorders and opioid use disorders			Rats	108	D	✓	✓	✓		✓	
Preclinical assessments for a new alcohol use disorders pharmacotherapy			Rats	2880	D	✓					

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
Pharmacology of synthetic cannabinoid-like drugs of abuse	Redacted by agreement		Mice Rats	720 1395	C, D	✓				✓	
Effects of striatal signaling in feeding behavior and metabolism			Mice Rats	120 540	C, D	✓				✓	
Simultaneous optical calcium imaging and functional MRI to study neural activity in the medial prefrontal cortex of rats			Rats	112	D	✓	✓			✓	
Generation of primary neuronal and glial cultures for evaluating novel viral vectors			Rats	144	C					✓	
Ensembles communication in motivational circuits			Mice	1290	D	✓					
Monitoring cellular stress in response to a high fat diet			Rats	336	D	✓					

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
Maintenance of animals not assigned to a research ASP	Redacted by agreement		Mice Rats Rhesus Squirrel monkey	7500 2400 10 100	C						
Electrophysiological Characterizations of Ventral Tegmental Area Glutamate and GABA neurons			Mice Rats	48818 11003	C, D	✓		✓		✓	
Holding protocol for genetically-modified rats and mice			Mice Rats	4320 25920	C, D					✓	
Transcytosis of nanoparticles in rat brain			Rats	328	D	✓					
Molecular alterations in context-induced reinstatement of methamphetamine seeking			Rats	320	D	✓	✓	✓		✓	
Neural mechanisms of relapse to aggression seeking in aggressive and non-aggressive mice			Mice	2088	D, E	✓				✓	

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
Molecular alterations in distinct ensembles associated with cocaine or heroin drug-seeking	Redacted by agreement		Rats	380	D	✓		✓		✓	
Distribution and insertion of silica microspheres into rat neocortex neurons			Rats	144	D	✓				✓	
Functional alterations in conditioned place preference			Mice	840	D	✓				✓	
Role of norepinephrine in the development of addiction-like symptoms in rats and mice			Mice Rats	585 472	C, D, E	✓	✓			✓	
Incubation of discriminative stimulus-induced cocaine craving			Rats	3168	D	✓	✓	✓		✓	✓
Neural mechanisms underlying the incubation of			Rats	384	D	✓					

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
oxycodone craving after cessation of oxycodone self-administration using a conflict procedure							✓			✓	
Neural mechanisms underlying incubation of prescription opioid craving and pain sensitivity	Redacted by agreement		Rats	792	C, D, E	✓	✓	✓		✓	✓
Electrochemical studies of brain glucose and oxygen in reward-related structures			Rats	180	D	✓				✓	
Context-induced reinstatement of oxycodone seeking in a rat model			Rats	812	D, E	✓	✓			✓	
Characterization of novel transgenic rats and adeno-associated viral (AAV) vectors			Rats	2016	D	✓				✓	✓
Transfer protocol for electrophysiological			Mice Rats		C						

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
and neurochemical recordings in rodent brain slices				180 180						✓	
Using neuromodulators and brain stimulation to block pain-induced opioid seeking	Redacted by agreement		Mice	1050	C, D, E	✓	✓			✓	
Holding protocol for transgenic mice			Mice	28020	C						
Nociceptive and anti-nociceptive circuits underlying opioid response during chronic pain			Mice	1065	D, E	✓	✓			✓	
The effects of transcranial magnetic stimulation (TMS) on motivated behaviors			Mice	4105	D	✓	✓	✓		✓	✓
Dissection of the drive/reward paradox			Mice	364	D	✓	✓			✓	

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
In vivo imaging and electrophysiological studies of striatal neuronal circuits during acute administration of tetrahydrocannabinol (THC)	Redacted by agreement		Mice	620	C, D	✓				✓	
Electrophysiological and neurochemical studies of cannabinoid actions in rodent brain slices			Mice Rats	834 150	C, D	✓				✓	
Studies on Sig1R-induced enddocanabinoids release from VTA dopamine neurons			Mice	912	D	✓				✓	
Role of neuropeptides and stress hormones in tolerance to alcohol and anxiety induced by withdrawal from alcohol			Rats	304	C, E					✓	

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
Monitoring endoplasmic reticulum stress in a chemogenic model of epilepsy	Redacted by agreement		Rats	550	D	✓				✓	✓
Characterization and studies of a novel rat model of HIV neuropathogenesis			Rats	17920	C, D	✓				✓	✓
Effect of chronic administration of naltrexone, buprenorphine or mifepristone on opioid-induced overdose death			Rats	276	D	✓				✓	
Role of anatomically and genetically defined cell populations in alcohol dependence			Rats	16512	D	✓				✓	
Effects of repetitive, low-intensity blast overpressure on			Rats	160	C, D	✓	✓			✓	

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
alcohol and heroin seeking in rats											
Monitoring endoplasmic reticulum (ER) calcium homeostasis using transgenic mice	Redacted by agreement		Mice	450	D	✓				✓	✓
Regulation of PCSK9 in the liver of alcohol dependent rats			Rats	288	D	✓	✓			✓	
The role of Ghrelin in diet-induced obesity in rats			Rats	288	E			✓		✓	
Neuropsychopharmacology of psychostimulant and opiod dependence			Rats	20976	D, E	✓	✓			✓	
Role of neuropeptides and stress hormones in alcohol dependence			Rats	11520	C, D	✓				✓	
Roles of cannabinoid and hormonal systems in drug addiction			Rats	2715	C, D	✓				✓	

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
Holding protocol linked to research protocols	Redacted by agreement		Mice	630	C, D					✓	
Technical development for MRI and Transcranial Magnetic Stimulation (TMS) on rodent animals			Mice	1331	D	✓	✓	✓		✓	
Consequences of adolescent nicotine exposure on subsequent nicotine dependence in adults: A Behavioral and fMRI Study			Rats	236	E	✓	✓	✓		✓	
Role of astrocyte activity in resting state functional magnetic resonance imaging signal: chemogenetics and fiber-photometry study			Rats	100	D	✓				✓	
The effects of transcranial magnetic			Rats	288	D						

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
stimulation (TMS) on rat brain										✓	
Evaluate the therapeutic efficiency of transcranial magnetic stimulation in cocaine addiction in rodents	Redacted by agreement		Rats	417	D	✓	✓	✓		✓	
Dissection of nucleus accumbens regulation of motivation and aversion			Mice	5380	C, D	✓	✓	✓		✓	
In vivo calcium imaging of social behavior in mouse models of neuropsychological disorders			Mice	3520	C, E	✓	✓			✓	✓
Characterization of Glutamine Synthetase Expression and Function in Oligodendrocytes			Mice	1972	C, D	✓				✓	
Assessment and surveillance of health			Mice Rats		C, D						

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
for quarantine rodents and research rodent colonies				1158 648						✓	
Basic biomethodology training for research staff	Redacted by agreement		Mice Rats	324 324	C						
Molecular mechanisms of incubation of methamphetamine and cocaine craving			Rats	288	D	✓	✓	✓		✓	
Modulatory roles of neurotransmitter systems in the reinforcing effects of new psychoactive substances in rodents			Mice Rats	2320 3230	C, D	✓	✓	✓			
Drug-induced molecular alterations within synapses			Rats	108	D	✓				✓	
Behavioural Flexibility and Decision Making in Rat Models			Rats	2992	D	✓	✓	✓		✓	
Role of endogenous cannabinoids in the			Rats		E						

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
ventral tegmental area on negative reinforcement		Redacted by agreement		165						✓	
The role of the lateral hypothalamus and ventral tegmental area in fear conditioning	Redacted by agreement		Rats	880	E	✓				✓	
Plasticity of somatodendritic excitability and signaling in monoaminergic brain circuits			Mice	1806	C, D, E	✓	✓			✓	
Postnatal Development of Glutamate-GABAergic projections to the Lateral Habenula in Rat			Rats	132	C					✓	
Fast scan cyclic voltammetry screening of candidate drugs for cocaine antagonist activity			Mice Rats	10580 12600	C, D	✓	✓	✓		✓	✓

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
Evaluation of the therapeutic efficacy of transcranial magnetic stimulation in opioid addiction in mice	Redacted by agreement		Mice	72	C					✓	✓
MicroPET scanning for rodents			Mice	196	D			✓		✓	✓
Neuromodulation on compulsive drug self-administration in rats			Rats	210	D	✓		✓		✓	✓
Nicotine vapor chamber efficacy: a behavioral and neuroimaging investigation			Rats	598	D, E	✓				✓	
Pharmacology of novel synthetic opioid drugs of abuse			Rats	1380	C, D	✓				✓	
Neuron glial interactions in the developing, mature, and aging basal ganglia			Mice	1958	C, D	✓				✓	✓

Appendix 5: Animal Usage

Project/Protocol Title	IACUC/OB Number	Principal Investigator	Species	Total Number of Animals Approved	Pain & Distress Category (1)	Special Considerations (use checkmark if applicable)					
						SS (2)	MSS (3)	FFR (4)	PR (5)	HAU (6)	NCA (7)
Transgenic rodent breeding and colony maintenance	Redacted by agreement		Mice Rats	99000 39000	C, D	✓				✓	

(1) If applicable, please provide a description / definition of any pain/distress classification used within this Appendix in the space below. If pain/distress categories are not used, leave blank.

(2) Survival Surgery (SS)

(3) Multiple Survival Surgery (MSS)

(4) Food or Fluid Regulation (FFR)

(5) Prolonged Restraint (PR)

(6) Hazardous Agent Use (HAU)

(7) Non-Centralized Housing and/or Procedural Areas (NCA), i.e., use of live animals in any facility, room, or area that is not directly maintained or managed by the animal resources program, such as investigator laboratories, department-managed areas, teaching laboratories, etc.

Pain/Distress Classification Description/Definition, if applicable:

These classification descriptions are used in Appendix 9, Animal Study Proposal form, Section H:
C - Minimal, Transient, or No Pain or Distress
D - Pain or Distress Relieved By Appropriate Measures
E - Unrelieved Pain or Distress

In the Table below, provide an approximate annual usage for all species:

Animal Type or Species	Approximate Annual Use
Mice	34,559
Rats	18,452
Nonhuman primates	48

Animal Type or Species	Approximate Annual Use

[Create additional rows by pressing TAB in the bottom-right box.]

Appendix 6: Personnel Medical Evaluation Form

Provide a ***blank*** copy of form(s) used by medically-trained personnel to review individual health assessment, individual risk assessment, health history evaluation, health questionnaire, periodic medical evaluation, etc. If form(s) are not used, include a description of how such evaluations are performed in the Program Description (Section 2.I.A.2.b.ii.1).d), Section 2 (Description). I (Animal Care and Use Program). A (Program Management). 2 (Personnel Management). b (Occupational Health and Safety or Personnel). ii (Standard Working Conditions and Baseline Precautions). 1) (Medical Evaluation and Preventive Medicine for Personnel). d).

Forms are not used. To support evaluation of the general description provided in section 2.I.A.2.b.ii.**1).e.** of the Program Description, the content of the enrollment medical evaluation, which is determined by the type of animals the worker will encounter at work and almost never includes a physical exam, is described in 2.I.A.2.b.ii.**1).e.** but outlined below:

1. An occupational and personal medical history which includes a review of:
   a. The functional demands and environmental factors associated with the proposed position;
   b. The type of animal(s) contacted;
   c. Other potential worksite health hazards; and
   d. The individual's medical history, including occupationally-indicated immunizations.
2. Counseling and informational handouts:
   a. Counseling includes:
      i) The importance of utilizing Standard Precautions;
      ii) The proper use of personal protective equipment;
      iii) Recommended first aid procedures;
      iv) The necessity to report all work-related injuries and illnesses, including allergies to OMS (see Section VII.B.3); and
      v) The process for activating the emergency medical response system.
   b. The worker is provided information and handouts regarding allergic reactions to laboratory animals (refer to the OMS Laboratory Animal Allergies procedure for more information) and relevant zoonoses based upon the animals used at the worksite.
3. Tetanus immunization. The worker is offered a booster dose of tetanus, diphtheria, and acellular pertussis (Tdap) vaccine if a dose of Tdap vaccine has not been administered as an adult, regardless of when the last dose of when the last dose of tetanus diphtheria (Td) vaccine was administered. The worker is offered a booster dose of Td vaccine, if the worker received a Tdap booster as an adult and ten or more years have lapsed since the last tetanus booster (see the OMS Tetanus, Diphtheria, Pertussis Immunization procedure for additional details.)
4. Documentation. The details of the enrollment are recorded both in the worker's OMS clinical record and the OMS Clinical Access Manager (CAM). This information in CAM is utilized for compliance reports and notifications to program enrollees.

Appendix 7: IACUC/OB Membership Roster

Please provide a Committee roster, indicating names, degrees, membership role, and affiliation (e.g., Department/Division).

Member Name	Lab	Position Title	PHS Policy Membership Role
Hoffman, Alexander, Ph.D.	CNRB, Electrophysiology Section	Staff Scientist	Chair
Stump, Kyle, DVM	OSD, Animal Care Section	Animal Program Director	Attending Veterinarian
Redacted by agreement			

Appendix 7: IACUC/OB Membership Roster

Redacted by agreement

Appendix 8: IACUC/OB Minutes

Please provide the latest two Minutes of the IACUC/OB meetings.

NIDA IRP ACUC MINUTES
24 May 2018

Redacted by agreement

1:00 PM

Proof of Quorum:

Present or [Absent]	A. Hoffman	K. Stump	
	Redacted by agreement		Redacted by agreement

*non-voting

**Alternate

***Guest

Appendix 8: IACUC/OB Minutes

A. **Announcements/New Business**

1. The ACUC approved the minutes 26 April 2018 meeting.

2. The committee reviewed the proposed Guideline for Assigning Pain and Distress Categories in Research Animals at NIDA-IRP and had some comments. It will be revised and reviewed again.

3. The committee unanimously approved the Transient Visitor and Breeding policies.

4. Redacted by agreement reported on the recent PAV visit. The PAV team watched Redacted by agreement prepare brain slices under the auspices of protocol Redacted by agreement There were no specific concerns with protocol adherence. During the discussion, it was manifested that Redacted by agreement is using isoflurane for the incisional paw procedure and that there was no method of euthanasia for animals that are dropped from the study outlined in the protocol. Redacted by agreement submitted a modification to the protocol to address these items.

B. Review of Initial Submissions, Renewals and Modifications (Significant Changes- Category 1):

1. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

2. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

3. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

4. (b)(5)

After careful deliberation, the Committee voted to table this protocol until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

Appendix 8: IACUC/OB Minutes

(b)(5)

(b)(5)

5. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

6. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

a. (b)(5)

(b)(5)

7. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

8. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

(b)(5)

9.

(b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

(b)(5)

10. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

Appendix 8: IACUC/OB Minutes

(b)(5)

C. Annual Reviews – all were approved

1. Redacted by agreement Electrophysiological and neurochemical studies of cannabinoid actions in rodent brain slices (Year1)
2. Redacted by agreement Studies on Sig1R-induced endocannabinoids release from VTA dopamine neurons (Year 1)
3. Redacted by agreement *In vivo* imaging of dorsal striatal circuits during the acquisition and consolidation of a motor learning skill (Year 2)
4. Redacted by agreement Hypothalamic neuronal circuits controlling motivated behaviors (Year 2)
5. Redacted by agreement Connectivity of hypothalamic neuronal circuits controlling motivated behaviors (Year 2)
6. Redacted by agreement *In vivo* imaging and electrophysiological studies of hypothalamic neuronal circuits controlling motivated behaviors (Year 2)
7. Redacted by agreement Nociceptive and anti-nociceptive circuits underlying opioid response during chronic pain (Year 1)

Appendix 8: IACUC/OB Minutes

D. Report of Significant Changes – Category 2 (Information only)

1. **Administrative Conditional (Category 2.1)**

Redacted by agreement Behavioral Flexibility and Decision Making in Rat Models
Modification Three – Additional animals, strain and procedure
Approved 04/19/2018

2. **Administrative Conditional (Category 2.2)**

a. none

E. Report of Minor Changes (Unconditional Administrative – Category 3): (Information only)

1. Redacted by agreement Behavioral Flexibility and Decision Making in Rat Models
Modification Two – Addition of personnel
Approved 04/17/2018

2. Redacted by agreement Role of norepinephrine in the development of addiction-like symptoms in rats and mice
Modification Four – Addition of personnel
Approved 05/11/2018

F. Report of Designated Member Reviews (follow-up from previous Full Committee Review) (Informational only)

Appendix 8: IACUC/OB Minutes

1. Redacted by agreement Molecular mechanisms of incubation of cocaine and methamphetamine craving
   Renewal of Redacted by agreement
   Approved 04/06/2018

2. Redacted by agreement Neural mechanisms underlying the incubation of oxycodone self-administration using a conflict procedure
   Modification Two – Add personnel, animals and experiments
   Approved 05/09/2018

3. Redacted by agreement Effects of striatal signaling in feeding behavior and metabolism
   Modification Three – Additional staff, animals and experiments
   Approved 05/09/2018

G. Report of Designated Member Reviews (outside of Full Committee Review) (Information only)

1. None.

NIDA IRP ACUC MINUTES
28 June 2018
Redacted by agreement
1:00 PM

Proof of Quorum:

Present or [Absent]	A. Hoffman	K. Stump	Redacted by agreement
	Redacted by agreement		

Appendix 8: IACUC/OB Minutes

Redacted by agreement

*non-voting
**Alternate
***Guest

A. **Announcements/New Business**

1. The ACUC approved the minutes 24 May 2018 meeting pending minor changes to the attendance roster.

2. Redacted by agreement gave the following PAV reports:
   a. Redacted by agreement rodent handling training – no issues noted.
   b. Redacted by agreement – the team watched the PI prepare the animals for behavioral box training the following issues were noted:

      The animals are being floor-fed which is not in the protocol. Redacted by agreement was not wearing gloves while handling the animals. Redacted by agreement stated the use of Redacted by agreement for longer than 12 hours which is not indicated in the protocol.

3. The committee approved the Animal Facility Emergency and Disaster Response Plan, Summer Students and NHP Policy, Pain Category SOP, General Conduct Within Animal Use Areas SOP and the Animal Restraint Policy pending minor modifications.

4. The committee approved the following SOPs: Animal Transport Vehicle SOP, Corridor and Clean Storage Areas SOP, Procurement for Feed, Bedding and Chemicals SOP, Procurement for Services, Supplies & Equipment SOP, Work Order Requests SOP and Personal Protective Equipment SOP.

5. Dr. Hoffman reminded the committee the Redacted by agreement approves the scientific merit of Non-human Primates.

B. **Review of Initial Submissions, Renewals and Modifications (Significant Changes- Category 1):**

Appendix 8: IACUC/OB Minutes

1. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

2. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

Appendix 8: IACUC/OB Minutes

(b)(5)

(b)(5)

3. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

(b)(5)

4. (b)(5)

Appendix 8: IACUC/OB Minutes

Renewal of (b)(5)

After careful deliberation, the Committee voted to table this protocol until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

(b)(5)

5. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

6. (b)(5)

After careful deliberation, the Committee voted to table this modification until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

(b)(5)

7. (b)(5)

After careful deliberation, the Committee voted to table this protocol until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

Appendix 8: IACUC/OB Minutes

(b)(5)

(b)(5)

8. (b)(5)

After careful deliberation, the Committee voted to table this protocol until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

Appendix 8: IACUC/OB Minutes

(b)(5)

(b)(5)

9. (b)(5)

After careful deliberation, the Committee voted to table this protocol until changed (as per the below list), at which time the protocol can be considered for approval by the designated member review process.

(b)(5)

(b)(5)

The annual reviews were approved.

None of the late agenda items were reviewed.

C. Annual Reviews – all were approved

1. Redacted by agreement Characterization of glutamate synthetase expression and function in oligodendrocytes (Year 1)
2. Redacted by agreement Preclinical assessments for a new alcohol use disorders pharmacotherapy (Year 1)
3. Redacted by agreement Regulation of PCSK9 in the liver of alcohol dependent rats (Year 1)
4. Redacted by agreement Role of neurotransmitters in alcohol dependence (Year 1)
5. Redacted by agreement Neuropsychopharmacology of psychostimulant and opioid dependence (Year 1)
6. Redacted by agreement Neurochemical correlates of addictive and anti-addictive drug actions using microdialysis and optogenetics (Year 2)

D. Report of Significant Changes – Category 2
(Information only)

1. **Administrative Conditional (Category 2.1)**

a. Redacted by agreement Regulation of PCSK9 in the liver of alcohol dependent rats
Modification Two – Additional experiment
Approved 05/22/2018

b. Redacted by agreement Incubation of discriminative stimulus-induced drug craving
Modification Three – Addition of test compound in the same class as currently approved
Approved 05/31/2018

2. **Administrative Conditional (Category 2.2)**

a. Redacted by agreement Incubation of discriminative stimulus-induced drug craving
Modification Four – Addition of strains
Approved 06/15/2018

E. Report of Minor Changes (Unconditional Administrative – Category 3):
(Information only)

1. Redacted by agreement Regulation of PCSK9 in the liver of alcohol dependent rats
Modification One – Addition of personnel
Approved 05/16/2018

2. Redacted by agreement Behavioral Flexibility and Decision Making in Rat Models
Modification Four – Addition of personnel
Approved 06/04/2018

3. Redacted by agreement Evaluation of the involvement of glycine receptors on opioid self-administration in male and female mice
Modification Four – Add rooms and personnel
Approved 06/04/2018

4. Redacted by agreement Role of neurotransmitters in alcohol dependence
Modification Two – Addition of personnel
Approved 06/04/2018

5. Redacted by agreement Neuropsychopharmacology of psychostimulant and opioid dependence
Modification Three – Addition of personnel
Approved 06/04/2018

6. Redacted by agreement Neural mechanisms underlying incubation of prescription opioid craving and pain sensitivity
Modification Six – Addition of personnel
Approved 06/06/2018

7. Redacted by agreement Neuromodulation on compulsive drug self-administration in rats
Modification One – Addition of personnel
Approved 06/07/2018

8. Redacted by agreement Postnatal Development of Glutamate-GABAergic projections to the Lateral Habenula in Rat
Modification One – Addition of personnel
Approved 06/07/2018

9. Redacted by agreement Nicotine vapor chamber efficacy: a behavioral and neuroimaging investigation
Modification One – Addition of personnel
Approved 06/08/2018

10. Redacted by agreement The effects of Transcranial Magnetic Stimulation (TMS) on rat brain
Modification Two – Addition of personnel
Approved 06/08/2018

11. Redacted by agreement Using neuromodulators and brain stimulation to block pain-induced opioid seeking
Modification Two – Addition of personnel

Approved 06/13/2018

12. Redacted by agreement Neurochemical correlates of addictive and anti-addictive drug actions using microdialysis and optogenetics
Modification Seven – Addition of personnel
Approved 06/14/2018

13. Redacted by agreement Context-induced reinstatement of oxycodone seeking in a rat model
Modification Five – Addition of personnel
Approved 06/15/2018

14. Redacted by agreement Neural mechanisms underlying incubation of prescription opioid craving and pain sensitivity
Modification Seven – Addition of personnel
Approved 06/15/2018

15. Redacted by agreement Neural mechanisms underlying the incubation of cocaine craving after cessation of oxycodone self-administration using a conflict procedure
Modification Two – Addition of personnel
Approved 06/15/2018

16. Redacted by agreement Plasticity of somatodendritic excitability and signaling in monoaminergeric brain circuits
Modification One – Addition of personnel
Approved 06/18/2018

17. Redacted by agreement Fast scan cyclic voltammetry screening of candidate drugs for cocaine antagonist activity
Modification One – Addition of personnel
Approved 06/18/2018

18. Redacted by agreement Microdialysis screening of candidate drugs for cocaine antagonist activity
Modification Three – Addition of personnel

Approved 06/15/2018

F. Report of Designated Member Reviews (follow-up from previous Full Committee Review)

(Informational only)

1. Redacted by agreement Nicotine vapor chamber efficacy: a behavioral and neuroimaging investigation
   Initial Submission
   Approved 05/22/2018

2. Redacted by agreement Developing a mouse oral fentanyl self-administration model for transcranial magnetic stimulation
   Initial Submission
   Approved 05/24/2018

3. Redacted by agreement Neuromodulation on compulsive drug self-administration in rats
   Initial Submission
   Approved 05/31/2018

4. Redacted by agreement Neural circuits of motivation and reward in mice and rats
   Modification 14 – Additional animals and experiments
   Approved 06/08/2018

5. Redacted by agreement Function study of the VTA afferents from parabrachial nucleus (PBN) or pedunculopontine tegmental nucleus (PPTg)
   Modification 10 – Additional animals and change of PI
   Approved 06/05/2018

6. Redacted by agreement Neural mechanisms underlying incubation of prescription opioid craving and pain sensitivity
   Modification Five – Additional animals and experiments

Approved of 06/05/2018

7. Redacted by agreement Context-induced reinstatement of oxycodone seeking in a rat model
Modification Four – Additional animals and experiments
Approved 06/05/2018

8. Redacted by agreement Plasticity of somatodendritic excitability and signaling in monoaminergic brain circuits
Initial Submission
Approved 06/08/2018

9. Redacted by agreement The Role of Ghrelin in Diet-Induced Obesity in Rats
Modification One – Additional experiments
Approved 06/08/2018

10. Redacted by agreement Role of Sigma-1 receptor in neuropathic pain
Modification Two – Additional rats and timepoints
Approved 06/08/2018

11. Redacted by agreement MicroPET scanning for squirrel monkeys
Initial Submission
Approved 06/13/2018

G. Report of Designated Member Reviews (outside of Full Committee Review)
(Information only)

1. None.

Appendix 9: IACUC/OB Protocol Forms

Please attach a ***blank*** copy of form(s) used by the IACUC/OB to review and approve studies. Include forms used for annual (or other periodic) renewal, modifications, amendments, etc., as applicable.

The text for the NIDA Animal Study Proposal (ASP) Form used for initial submissions, renewal, and modification follows:

Protocol #
APPROVAL DATE
EXPIRATION DATE

NATIONAL INSTITUTES OF HEALTH

NIDA - IRP

ANIMAL STUDY PROPOSAL

Click here **for instructions for completion of the form. Click** here **for instructions for modifying an existing protocol.**

For submission mail to: NIDAIRPACUC@mail.nih.gov

A. ADMINISTRATIVE DATA:

Institute, Center, or Division: NIDA IRP

Principal Investigator: _____

Building/Room: _____ *Telephone* (443) 740-_____

Branch: _____

Project Title: _____

Appendix 9: IACUC/OB Protocol Forms

[ ] Initial Submission

[ ] Renewal of ASP Proposal number _____

[ ] Modification ____(eg 1,2,3...)

[ ] Change of PI only

*List the names of all individuals (**including the PI**) authorized to conduct procedures involving animals under this proposal.*

Name	Type of animal contact:		Procedures performed (one procedure and competency determination per line):	Fully Qualified	If not qualified trained to full competency by:	IACUC Verification		
	NHP*	Rodent				AEP	NIH Course	Questionnaire

*Use of awake/unanesthetized NHPs requires Awake_NHP_Procedure_Training certification.

B. ANIMAL REQUIREMENTS:

Species: __

Age/Weight/Sex Unit:__

Stock, Strain and/or Genotype: __

Source(s): ___

Holding Location(s): ____

Number of Animals by species for initial submission:

Year 1	Year 2	Year 3	= Total

Copy table to add animals by modification.

If animals are transgenic, are there any known associated deleterious phenotypes? Yes [ ] No [ ]
If yes, please describe them in Section F, Alternative Endpoints.

If animals are moved to other locations, list each animal procedure and testing area according to the duration the animal is in the area.

Duration in non-holding location	List locations		
For longer than 24 hrs†			
For 12 to 24 hrs			
For less than 12 hrs			

† *Non-standard housing for longer than 24 hrs requires following the Animal Testing Within the Animal Facility For Longer Than 24 Hours , or the Satellite Rodent Housing SOP for animals housed outside the Animal Facility.*

C. TRANSPORTATION: *Transportation of animals must conform to all NIH and Facility guidelines/policies. If animals will be transported between buildings, through clinical areas or to another facility, describe the methods and containment to be utilized. Transportation within a single NIDA building does not need to be described here.*

[ ] Not Applicable

D. STUDY OBJECTIVES: *Briefly explain in non-technical terms the aim of the study and why the study is important.* ***THIS SECTION IS MEANT TO BE UNDERSTANDABLE BY AN 8th GRADE STUDENT.***

E. RATIONALE FOR ANIMAL USE:

1) Explain your rationale for animal use.

2) Justify the appropriateness of each species selected.

3) Justify the number of animals to be used by explaining group size(s) determination.

F. DESCRIPTION OF EXPERIMENTAL DESIGN AND ANIMAL PROCEDURES:

1. Description of Experimental Design and Animal Procedures:

   Briefly explain the experimental design and specify all animal procedures. This description should allow the ACUC to understand the experimental course of an animal from its entry into the experiment to the endpoint of the study. Remember, approved proposals are subject to release to the public under the Freedom of Information Act. DO NOT include proprietary or classified information on the ASP form. Flow charts, time lines, etc. may be helpful to demonstrate how different parts of the study are related. If the ACUC cannot visualize each of the procedures being performed on the animals, it cannot properly discharge its responsibility. Details of each procedure that may impact on the pain or stress potential of that procedure should be specified. Specifically address the following in your description:

   - *Injections or Inoculations (substances, e.g., infectious agents, adjuvants, etc.; dose, sites, volume, route, and schedules)*
   - *Blood Withdrawals (volume, frequency, withdrawal sites, and methodology)*
   - *Non-Survival Surgical Procedures (Provide details of survival surgical procedures in Section G.)*
   - *Radiation (dosage and schedule)*
   - *Methods of Restraint (e.g., restraint chairs, collars, vests, harnesses, slings, etc.)*
   - *Animal Identification Methods – (Include an individual marking method in addition to cage cards, e.g., ear tags, tattoos, collar, , etc. and cage card)*
   - *Other Procedures (e.g., survival studies, tail biopsies, etc.)*

- *Resultant Effects, if any, the animals are expected to experience (e.g., pain or discomfort, ascites production, etc.)*

2. Alternative endpoints:

   - *Define precisely the humane endpoints including the assessment criteria (i.e., tumor size, percentage body weight gain or loss, inability to eat or drink, behavioral abnormalities, clinical symptomatology, or signs of toxicity) when the administration of tumor cells, biologics, infectious agents, radiation or toxic chemicals are expected to cause significant symptomatology or are potentially lethal.*
   - *Provide the frequency of animal observations*
   - *Identify the person(s) responsible for assessment and recognition of the alternative endpoints*
   - *Indicate actions taken upon reaching the alternative endpoint.*

3. Pharmaceutical Grade Compounds:

If administering compounds to animals, please indicate the quality of compounds by checking the appropriate box below:

__ All compounds including their vehicles are pharmaceutical grade (no need to fill in chart).

__ At least some compounds administered to animals are not pharmaceutical grade. Fill in chart below for a required scientific justification for all compounds on line 1, and for any individual compounds having different justifications on subsequent lines.

Non-Pharmaceutical Grade Compound	**Grade, Quality, or Purity (see e.g. below)**	**Non-pharmaceutical grade compounds are used in these studies because (check those that apply):**			
		Pharmaceutical grade and alternatives not available	**Direct comparison with previous results required**	**Greater concentration or purity required than that available**	**Pharmaceutical formulation is not scientifically acceptable**
1)					
2)					

3)					
4)					
5)					

Other **scientific** justification (please describe): _____

Definitions:

Pharmaceutical-grade compound: Any active or inactive drug, biologic, reagent, et cetera, which is approved by the FDA for which a chemical purity standard has been written or established by any recognized pharmacopeia.

Pharmacopeia: A book or a compendia, such as the US Pharmacopeia [USP], the National Formulary [NF], the British Pharmacopoeia [BP], the Pharmacopoeia of the Council of Europe [EP]. See the combined standards of the USP and the NF.

Analytical grade: ~99% purity (typically has an available Certificate of Analysis). For the purposes of ASP approval, the NIDA-IRP ACUC considers all compounds that are of equal or superior quality and/or purity to that of pharmaceutical grade as "analytical grade." Such designations include "Extra Pure Grade," "reagent grade" (with any of its various adjectives), "chemically pure grade," "biotech grade," "biochemistry grade," and "ultra pure grade."

New investigational compound: Supplied by its manufacturer for testing in an experimental setting only with or without chemical purity standards.

Refer to ARAC Guidelines for the Use of Non-Pharmaceutical Grade Compounds in Laboratory Animals for further information

All non-pharmaceutical grade drugs must be prepared in accordance with the NIDA SOP or by a compounding pharmacist.

G. SURVIVAL SURGERY: *If proposed, complete the following:*

[ ] Not Applicable

1. *Identify and describe the surgical procedure(s) to be performed. Include the aseptic methods to be utilized.*

2. *Who will perform surgery and what are their qualifications and/or experience*?

Name	Procedures	Fully qualified in all relevant animal procedures	Will be trained and supervised by:	IACUC Verification	
				Yes	No

3. *Where will surgery be performed, Building and Room*?

4. *Describe post-operative care including use of analgesics, the recording of at least three days of health observations on post-operative cage cards,and identify the responsible individual. Any observed post-operative complications must be reported to the veterinarian.*

5. *Has survival surgery been performed on any animal prior to being placed on this study? Y/N___ If yes, please explain*:

6. *Will more than one survival surgery be performed on an animal while on this study? Y/N___ If yes, please justify*:

H. PAIN OR DISTRESS COLUMN: *The ACUC is responsible for applying U.S. Government Principle IV. Contained in Appendix 3: "Proper use of animals, including avoidance or minimization of discomfort, distress, and pain when consistent with sound scientific practices, is imperative. Unless the contrary is established, investigators should consider that procedures that cause pain or distress in human beings might cause pain or distress in other animals." Check the appropriate column(s) and indicate the approximate number of animals in each. Sum(s) should equal total from Section B.*

If rats or mice are indicated in Column E, a scientific justification is required in section F to explain why the use of anesthetics, analgesic, sedatives or tranquilizers during and/or following painful or distressful procedures is contraindicated. If monkeys are indicated in Column E, attachment two must be completed. Note: the ASP is subject to the Freedom of Information Act.

Number of Animals Used Each Year for initial submission:

	Year 1	Year 2	Year 3
USDA Column C - *Minimal, Transient, or No Pain or Distress*			

USDA Column D - *Pain or Distress Relieved By Appropriate Measures*			
USDA Column E - *Unrelieved Pain or Distress*			

Copy table to add animals by modification.

Describe your consideration of alternatives to procedures listed for Column D and E that may cause more than momentary or slight pain or distress to the animals, and your determination that alternatives were not available. [Note: Principal Investigators must certify in Section N # 5 that no valid alternative was identified to any described procedures which may cause more than momentary pain or distress whether it is relieved or not.] Delineate the methods and sources used in the search below. Database references must include the databases searched, the date of the search, period covered, and keywords used.

At a minimum the following terms must be searched:

animal testing alternatives OR animal use alternatives OR animal experimentation AND (the proposed painful or distressful procedures separated by OR if multiple procedures). Please provide your keywords/search strategy using the applicable Boolean operators; AND, OR and NOT.

Example strategy: animal use alternatives OR animal experimentation OR animal testing alternatives AND microdialysis OR intracranial cannulation OR intravenous catheterization

Also, see Database Searching for Alternatives to Painful Procedures *for guidance regarding the literature search requirement for alternatives to distressful or painful procedures*

Date(s) of search	
Years covered by search	
Databases searched/sources consulted	
(If using personal resources (e.g. subject-expert	

consultants), state who was contacted, when they were contacted and their level of expertise in the subject area)	
Key words presented as search strategy(ies)	
Narrative	

I. ANESTHESIA, ANALGESIA, TRANQUILIZATION: *For animals indicated in Section H, Column D, specify the anesthetics, analgesics, sedatives or tranquilizers that are to be used (including topicals) in the appropriate tables below. The use of analgesics and anesthetics must be in accordance with the NIDA IRP ACUC Guidelines Analgesia Guidelines and Anesthesia Guidelines.*

[ ] Not Applicable

Anesthesia

Drug Name (use a separate row for boosters)	Procedure	Dose (mg/kg)	Route	Duration	Concentration or Volume

If using isoflurane, state the scavenging method:					

Analgesics/Tranquilizers

Drug Name	Procedure	Dose (mg/kg)	Route	Frequency or Schedule	Concentration or Volume

J. METHOD OF EUTHANASIA OR DISPOSITION OF ANIMALS AT END OF STUDY: *Indicate the proposed methods of euthanasia for all animals in the study* ***including those removed from the study.*** *If a chemical agent is used, specify the dosage and route of administration. All methods must conform with the* AVMA Guidelines for the Euthanasia of Animals -2013 edition. *Indicate the method of carcass disposal if not as MPW.*

In addition::

All methods of euthanasia MUST include a physical method of euthanasia, either as the primary or secondary method. Physical methods commonly used include decapitation, exsanguination (perfusion), bilateral pneumothorax, or cervical dislocation (mice). If any animals will be euthanized by the Animal Care staff upon request, then indicate which animals and state that they will be euthanized by carbon dioxide exposure and a physical method by the Animal Care staff in accordance with the NIDA SOP.

K. HAZARDOUS AGENTS:

The use of potentially hazardous materials requires DOHS, IBC, and/or DRS (as appropriate) pre-approval. The appropriate Registration Document numbers are required to be listed. Actual registration documents and material safety documents may be required by the ACUC as an attachment. The use of hazardous chemicals requires a comprehensive chemical risk assessment for work on this ASP, performed by the Investigator and a DOHS Safety Representative. If an irradiator is to be used, then all individual users must comply with

radiation safety requirements for irradiator training, and all individual assessors must comply with applicable security requirements for escorts and proximity card access approval. If no hazards are being proposed then the "No" box needs to be checked.

Hazardous Material	YES	NO	List Agents	Registration Document
Radionuclides				
Biological Agents				PRD#:
Recombinant DNA				RD#:
Chemicals/Experimental Drugs				

Refer to the NIH-IBC documentation for the specific biosafety practices and procedures for working with the listed hazardous biological material.

Study conducted at Biosafety Level: _____

1. List or refer to any applicable SOP's for all materials used:
2. Describe the practices and procedures required for the safe handling and disposal of contaminated animals and material associated with this study. Also describe methods for removal of radioactive waste and, if applicable, the monitoring of radioactivity.

L. BIOLOGICAL MATERIAL/ANIMAL PRODUCTS:

Biological material and animal products (cell lines, tissues, tumors) have been incriminated repeatedly as vehicles for the introduction of contagious organisms, e.g., ectromelia, LCM, and MHV. NIDA requires approval before the introduction of any rodent, rodent product, or biologicals that may harbor agents of animal diseases considered dangerous to the programs of NIDA. If it is not certain that the biological materials/animal products to be used have been produced and maintained in a manner which excludes/eliminates any potential pathogens they may be required to be tested before being transported to NIDA facilities.

[ ] Not Applicable

1. Specify Material
2. Source_____ Material Sterile or Attenuated ___Yes ___No

3. If derived from rodents, has the material been MAP/RAP/HAP/PCR tested?

___Yes (Attach copy of results) ___No ___N/A

4. I certify that the MAP/RAP/HAP/PCR tested materials to be used have not been passed through rodent species outside of the animal facility in question and/or the material is derived from the original MAP tested sample. To the best of my knowledge the material remains uncontaminated with rodent pathogens.

____Initials of Principal Investigator.

M. SPECIAL CONCERNS OR REQUIREMENTS OF THE STUDY: - *List any special housing, equipment, animal care (i.e., special caging, water, feed, or waste disposal, etc.). If holding animals in laboratories for more than 12 hours will be necessary, it should be described. Any location holding animals for more than 12 hours requires ACUC evaluation for compliance with Guide regulations. If surgically modified animals are needed for this study, i.e., adrenalectomized, splenectomized, etc., their surgical modification, source, and any special care requirements should be described.*

All rodents must be group housed unless this would have adverse effects over the course of the study. Single housing due to attrition must also be justified (see 1b, below).
See ARAC Guidelines for Social Housing for more information.

1. **Individual Housing:**

a. If housing individually for all or a portion of study, provide justification (e.g. to protect surgical sites or implants, need for operant chamber housing, microdialysis and/or monitoring food restriction) in the space below:

[ ] Not applicable.

b. If animals that are originally group housed become individually housed as a result of cage mate loss or removal, the animals will not be re-housed with other animals due to:

[ ] the introduction of variables potentially affecting the integrity of the study.
[ ] the animals will not be socially compatible with others of this age, strain and/or sex.

[ ] Not applicable.

2. **Special feeding/drinking conditions:**

Describe food/water restriction and monitoring. See Guidelines for Diet Control in Behavioral Animal Studies for guidance.

[ ] Not applicable.

3. **Rodents**: Home cage supplementation.

[ ] Provide supplementation:

Mice: [ ] Nestlets (preferred)

[ ] Igloos

[ ] Other _____

Rats: [ ] Paper tube (preferred)

[ ] Nylabone

[ ] Other _____

[ ] No supplementation

4. **Non-Human Primate Enrichment Plan**

Explain your environmental enrichment plan or indicate compliance with the environmental enrichment plan in the species-specific SOPs.

5. **Use of audio/video recording devices**

Indicate whether audio/video recordings are utilized with any of the procedures listed in the protocol. Note that the use of all audio/video recording devices must be in compliance with NIH Manual Policy 3047: https://policymanual.nih.gov/3047

[ ] Yes

[ ] No

If yes, summarize briefly:

N. PRINCIPAL INVESTIGATOR CERTIFICATIONS:

1. I certify that I have attended an approved NIH investigator training course.

   Most current year of training: ____

2. I certify that I have determined that the research proposed herein is not unnecessarily duplicative of previously reported research.
3. I certify that all individuals working on this proposal are participating in the NIH Animal Exposure Surveillance Program.
4. I certify that the individuals listed in Section A are authorized to conduct procedures involving animals under this proposal, have attended the course "Using Animals in Intramural Research: Guidelines for Animal Users", and received training in the biology, handling, and care of this species; aseptic surgical methods and techniques (if necessary); the concept, availability, and use of research or testing methods that limit the use of animals or minimize distress; the proper use of anesthetics, analgesics, and tranquilizers (if necessary); procedures for reporting animal welfare concerns.
5. ***FOR ALL CATEGORY D AND CATEGORY E PROPOSALS (see section N):***

I certify that I have reviewed the pertinent scientific literature and the sources and/or databases as noted in Section H, and have found no valid alternative to any procedures described herein which may cause more than momentary pain or distress, whether it was relieved or not.

6. I will obtain approval from the ACUC before initiating any significant changes in this study.

Principal Investigator: Signature_____ Date

O. CONCURRENCES: PROPOSAL NUMBER_____(leave blank)

Laboratory/Branch Chief certification of review and approval of initial submissions and renewals on the basis of scientific merit. (Scientific Director's signature required for proposals submitted by a Laboratory or Branch chief.)

Name _____
Signature_____ Date_____

Scientific Director certification of review and approval of initial submissions or renewals where the Principal Investigator is also the Branch Chief, and/or of initial submissions or renewals involving non-human primates,.

Name _____
Signature_____ Date_____

P. FINAL APPROVAL:

Safety Representative certification of review and concurrence. (Required of all studies utilizing hazardous agents.)

Name See attached Signature See Attached Date_____

Attending Veterinarian certification of review.

Name Kyle Stump, DVM, DACLAM Signature_____ Date_____

Appendix 9: IACUC/OB Protocol Forms

Certification of review and approval by the Animal Care and Use Committee.

CHAIRPERSON: Alex Hoffman, Ph.D. Signature_____ Date_____

Attachment 1 – Emergency POCs and Animal Disposition

	Primary Contact	Secondary Contact
Name		
Work phone number		
Home phone number		
Pager/Cell phone (if applicable)		
Home email (if applicable)		
Euthanasia (e.g. at vet discretion, list criteria for euthanasia, notify lab contact to perform.)		
Carcass disposition instruction (e.g. freeze, refrigerate, tissue harvest, instrumentation recovery)		
Potential or expected complications by species		
Treatment by complication (at vet discretion* or list specific		

treatments or drugs that are contraindicated)		

Phone numbers will be distributed to only essential personnel and access will be restricted to protect your private information.

***The veterinarian will take the appropriate action in an emergency if no response from the PI/P●C is received within a half an hour of the attempted notification**.

Attachment Two

Column E Explanation Form For Regulated Species (non-rodents)

Though Column E studies do involve pain and/or distress, properly selected endpoints are applied to minimize pain and/or distress and adverse effects to the animals while accomplishing the scientific goals.

This form is intended as an aid to completing the Column E explanation. Names, addresses, protocols, veterinary care programs, and the like, are not required as part of an explanation. A Column E explanation must be written so as to be understood by lay persons as well as scientists.

1. **Registration Number:** Redacted by agreement

2. **Number of animals used under Column E conditions in this study.**

3. **Species (common name) of animals used in this study.**

4. **Explain the procedure producing pain and/or distress, including reason(s) for species selected. (*from ASP Section F*)**

5. Provide scientific justification why pain and/or distress could not be relieved. State methods or means used to determine that pain and/or distress relief would interfere with test results *(from ASP, Section F).* Provide summary of supportive care measures (if applicable).

Signature of Principal Investigator:

Date:

Signature of Chairman, ICD-ACUC:

Date:

ARAC Guidelines

The text for the NIDA Annual Review Form follows:

NIDA - IRP

1st/2nd) ANNUAL REVIEW of an ANIMAL STUDY PROTOCOL

ADMINISTRATIVE DATA:

Institute, Center, or Division: NIDA IRP

Principal Investigator: ____

Building/Room: _____ *Telephone*

Appendix 9: IACUC/OB Protocol Forms

Branch: ___

Project Title: _____

To date, you have used ___ (animals) You have __ remaining.

1. Is this study Active?
   ☐ Yes (please Continue) ☐ No (Please complete item 4, sign and return this form. (The protocol be inactivated)

2. Are there any changes to this protocol not yet approved by an modification such as:

	Yes	No		Yes	No
Overall aims or objectives			Procedures performed on animals, e.g., drugs, methods, dosing		
Levels of Pain/distress			Method of euthanasia		
Surgical Procedures: from minor to major, non survival to survival or single to multiple			Use of biological materials / animal products		
Additional animals needed (greater than 10%)			Special housing or animal requirements (includes use or addition of audio/video equipment)		
Genus or Species			Lab holding location		
Change in PI or Personnel			Safety considerations		
Addition of hazardous Agents			Other changes not mentioned		

If you answered yes to any of these questions please explain and submit a modification to the protocol

3. If monkeys are used and single housed, does the justification for single housing still apply?
   ☐ Yes (please Continue) ☐ No (Please explain) ☐ N/A

4. Were any substantial unanticipated animal health-related difficulties or significant adverse events encountered in performing the described procedures?
   ☐ Yes (please explain) ☐ No

Principal Investigator: Signature_____ Date_

Approvals:

Kyle Stump, DVM, DACLAM Signature_____ Date _____

CHAIRPERSON:

Alex Hoffman, Ph.D. Signature_____ Date _____

Please make any necessary changes to the table and complete any highlighted areas.

	Primary Contact	Secondary Contact
Name		
Work phone number		
Home phone number		
Pager/Cell phone (if applicable)		
Home email (if applicable)		
Euthanasia (e.g. at vet discretion, list criteria for euthanasia, notify lab contact to perform.)		

Carcass disposition instruction (e.g. freeze, refrigerate, tissue harvest, instrumentation recovery)		
Potential or expected complications by species		
Treatment by complication (at vet discretion* or list specific treatments or drugs that are contraindicated)		

Phone numbers will be distributed to only essential personnel and access will be restricted to protect your private information.

***The veterinarian will take the appropriate action in an emergency if no response from the PI/POC is received within a half an hour of the attempted notification.**

Appendix 10: IACUC/OB Periodic Report

Please attached a copy of the latest facilities (including laboratory inspections) and program assessment report conducted by the IACUC/OB.

SEMIANNUAL REPORT

ANIMAL CARE AND USE
PROGRAM REVIEW AND FACILITY INSPECTION
OF THE

National Institute on Drug Abuse (NIDA)

APRIL 2018

Section A – Site Visits & Program Review

1) Inspections of the ***NIDA*** animal facilities (AF), satellite holding facilities (SF), USDA-defined study areas for regulated species (SA) and areas where any surgical manipulations (Surg) are performed (as applicable) were conducted as indicated below:

Location	Type	Date	ACUC Members
Redacted by agreement	AF, Surg	03/08/18	Redacted by agreement
	SF	03/08/18	Stump, Hoffman, Redacted by agreement
	AF, SA, Surg	03/07/18	Stump, Hoffman, Redacted by agreement Redacted by agreement

2) Visits by at least one member of the ACUC to all remaining areas where animal activities were performed were conducted. These visits occurred during the previous six months and findings and corrective actions are described in this or the previous semiannual report.

3) The following document(s) was/were used as the basis for review of the animal care and use program:

	Document/Resource:
✓	Guide for the Care & Use of Laboratory Animals, 8th Edition (Guide)
✓	AAALAC Program Description
	OACU "Animal Program Semiannual Assessment Checklist" (1 page summary)
	OACU/OLAW "Semiannual Program Review & Facility Inspection Checklist" (24 pages)
✓	Other documents/resources (please specify) Animal Welfare Act Regulations

✓	AVMA Guidelines for the Euthanasia of Animals: 2013 Edition

Additional Topic Review Dates and Topics:

Fall Program Review Date(s) & Topics	3/27/18	Create Expired Products Policy – draft review.	Discussion of PAV program frequency of visits.	Establishment of guidelines for Pain Category classification. Subcommittee created.	Discussion of changeover to electronic ASP form.	Addition of information on procedures performed in rooms for semiannual inspection data sheets.	Discussion of Radiation Safety Office receipt of ASPs using isotopes in animals. Guidelines established.	Discussion of providing for catheter flushing for animals on holding protocols.	Information shared from 2018 PRIM&R meeting.

4) The program review was conducted in the following manner:

	Program Review Process
	Full committee member review for *ALL* of the review, i.e. the documents/resources listed in A3) are included in the meeting packet and reviewed at a fully convened meeting
	Full committee and subcommittee review, i.e. the documents/resources listed in A3) are assigned to various members who review their parts/sections and then they discuss their reviews with the full committee for a final review/approval
	Designated member review, i.e. the documents/resources listed in A3) are assigned to various members who review their parts/sections and then report back to the full committee the results of their designated review
✓	Other, please describe: Subcommittee review, participation was opened to all ACUC members. The documents and resources listed in A3 were distributed to subcommittee members. Agenda items were requested for discussion, and these were reviewed at a convened meeting. A summary of the program review was

	presented to the full committee on 04/26/18.

Section B – Regulatory Compliance:
Except as noted in Sections E, F, and G below, the facilities and program are in full compliance with the Public Health Service Policy, the Animal Welfare Act Regulations and the Guide, which were used as the basis for this evaluation.

Section C – Program Changes:
The following administrative and procedural changes have occurred since the program was last evaluated:

1) Administrative/Procedural Changes: None

2) Key Personnel Changes - ACUC Chair, ACUC Attending Vet, APD, or Program Manager:

Role (ACUC Chair, ACUC AV, IC APD, or IC Animal Program Manager)	**Name**	**Action** (joined or departed)
Redacted by agreement		Departed
ACUC Chair	Alex Hoffman, Ph.D.	Joined

3) Animal Facility/Area Changes:

Facility Type (AF/SF)	**Location**		**Action** (opened, closed, under renovation, etc.)
Redacted by agreement	Redacted by agreement		Opened

Section D – Guide Departures & USDA Exceptions:
Departures from the standards of the *Guide* and exceptions to the USDA *Animal Welfare Act Regulations,* which have been approved by the Animal Care and Use Committee, include the following:

1. Departures from the Guide: **None**

2. Exceptions to the AWAR: **None**

Appendix 10: IACUC/OB Periodic Report

Section E – Previous Deficiencies & Plans:
The committee validated that the plans and schedules for deficiencies noted during the previous **NIDA** program review, and facilities and laboratory inspections were achieved within the time intervals projected on the previous semiannual report.

Section F – Current Deficiencies & Plans:
Deficiencies found *over the past 6 months* during **NIDA** program review, facility inspections, and laboratory inspections, are as follows:

	Deficiency	[1]M/S	Location	Correction Plan	Responsible Party	Scheduled Completion Date (mm/dd/yy)	[2]Status: C/P
1	(b)(5)						
2	Insufficient sanitation	M	Redacted by agreement	A plan for cleaning and sanitizing the cubicle doors and changing the air filters has been developed and implemented. A record of the cleaning and filter changing will be kept in the area.	PI	04/04/18	C
3	Non-sanitizable chair	M		The chair has been discarded.	PI	03/19/18	C
4	Vacuum with non-Hepa filter	M		The vacuum without a HEPA filter has been taken out of use.	PI	03/16/18	C
5	(b)(5)						

6	Incomplete post-surgical documentation	M	Redacted by agreement	The surgery follow-up cards have been completed. Post-Bac talked to and re-emphasized the importance of recording all post-operative observations after surgery.	PI	03/16/18	C
7	Charcoal scavenging canister over weight.	M		The used charcoal canister has been discarded and replaced by a new one. The lab staff has been re-trained regarding the weighing procedures for charcoal canisters.	PI	03/16/18	C
8	Expired saline	M		The item has been discarded and staff have been re-trained on proper labeling and discarding of solutions.	PI	03/16/18	C
9	Non-sterile vial containing drug.	M		This item has been discarded.	PI	03/16/18	C
10	Expired suture material	M		PI has discarded expired suture material and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	03/29/18	C
11	Expired saline	M		PI has discarded expired saline and retrained staff consistent with OLAW policy on the use of expired	PI	03/16/18	C

				substances in animals.			
12	Expired saline	M	Redacted by agreement	PI has discarded expired saline and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	03/15/18	C
13	Expired saline	M		PI has discarded expired saline and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	03/15/18	C
14	Expired saline	M		PI has discarded expired saline and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	03/19/18	C
15	Expired food reward pellets	M		PI has discarded expired food reward pellets and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	03/16/18	C
16	Expired food reward pellets	M		PI has discarded expired food reward pellets and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	03/29/18	C
17	Expired food reward pellets	M		PI has discarded expired food	PI	03/29/18	C

				reward pellets and retrained staff consistent with OLAW policy on the use of expired substances in animals.			
18	Expired medicated feed	M	Redacted by agreement	PI has discarded expired medicated feed and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	03/16/18	C
19	Expired drug	M		PI has discarded the expired drug and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	03/16/18	C
20	Expired drug	M		PI has discarded the expired drug and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	03/28/18	C
21	Expired drug	M		PI has discarded the expired drug and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	03/16/18	C
22	Expired drug	M		PI has discarded the expired drug and retrained staff consistent with OLAW policy on the use of expired	PI	03/15/18	C

				substances in animals.			
23	Expired drug	M	Redacted by agreement	PI has discarded the expired drug and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	3/28/18	C
24	Expired drug	M		PI has discarded the expired drug and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	3/30/18	C
25	Expired drug	M		PI has discarded the expired drug and retrained staff consistent with OLAW policy on the use of expired substances in animals.	PI	3/30/18	C

[1]M=minor; S=significant [2]C=corrected; P=pending

Section G – Reportable Events:

PHS Policy (i.e. OLAW) reportable events that occurred in the last 6 months or that are still awaiting final disposition are as follows:

SA 1st noted	Description of event	Current Status
11/23/15	Improper surgical procedures	Resolved 12/17
06/20/17	Improper euthanasia procedure	Resolved 12/17

Section H – Shared & Central Facilities:

This semiannual report also encompasses review and oversight of animals and animal activities which were present or occurred

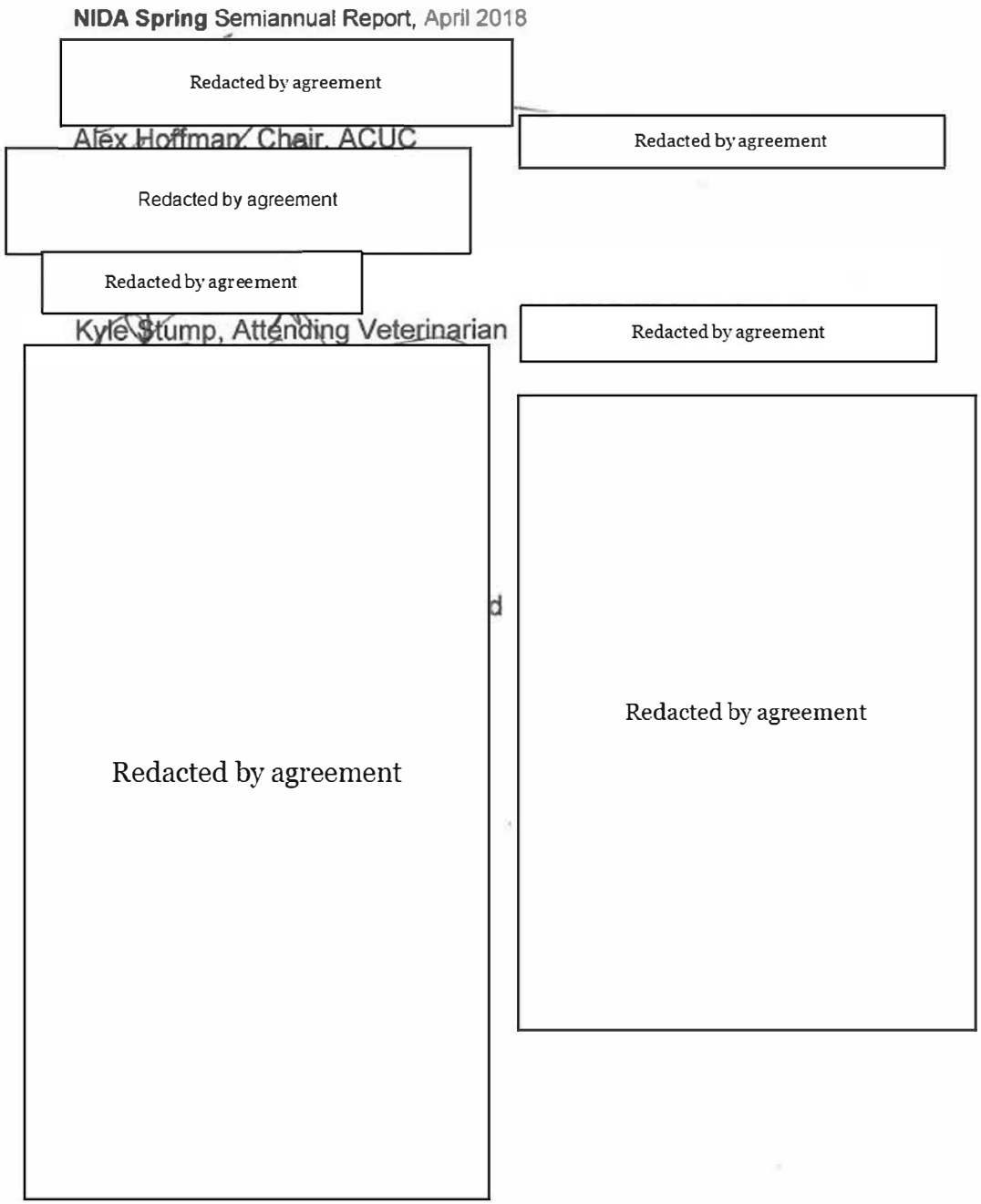
in shared or central facilities. Deficiencies were noted and transmitted directly to the facility, and if necessary, to the responsible Animal Care and Use Committee. These reviews were conducted as indicated below:

Building	Date	ACUC Members	
Redacted by agreement	03/26/18	Redacted by agreement	Stump

Section I – Minority Report

There is not a minority report filed with this semiannual report.

Appendix 10: IACUC/OB Periodic Report



Appendix 10: IACUC/OB Periodic Report

Semiannual Report Attachment 3
Supplemental Information
Spring 2018

Instructions: Submit the following information with your Spring 2018 Semiannual Report as a separate file called IC-SI-S18.

Performance Standards:
Provide a description of ACUC approved performance standards. *For additional information and examples, see the "Guide Departures & Performance Standards" document developed by OACU.*

In the ACUC protocol for NIDA's centralized breeding program, dams with litters < 5 days of age will forgo one weekly cage change so as not to distress the dam, and to avoid cannibalism. Therefore, cage changing may be delayed 7 days, for a maximum of 14 days between cage changes. Cages are monitored daily by animal care staff and changed if excessive moisture and/or feces are present. Fresh chow and water will always be provided.

Cage change frequency for rodents may be extended during a disaster response, as per the ACUC-approved NIDA Animal Facility Emergency and Disaster Response Plan. Animals will be observed daily by animal care staff as conditions permit. As soon as possible, cages will be changed and animal care staff will resume the regular cage changing schedule upon the Animal Program Director's decision that disaster conditions are mitigated.

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Summarize the heating, ventilation and air conditioning (HVAC) systems for each animal facility, ***including all satellite facilities***. Include ***all animal holding rooms*** (including satellite holding rooms), surgical facilities, procedure rooms, and support spaces integral to animal facilities (e.g., cage wash, cage and feed storage areas, necropsy, treatment).

Location/Building/Facility:	Redacted by agreement

In the text box below, provide a general description of the mechanical systems used to provide temperature, humidity and air pressure control. Include details such as:

- the source(s) of air and air recirculation rates if other than 100% fresh air
- treatment of air (filters, absorbers, *etc.*)
- design features such as centralized chilled water, re-heat coils (steam or hot water), individual room vs. zonal temperature and relative humidity control, the use of variable air volume (VAV) systems and other key features of HVAC systems affecting performance
- features that minimize the potential for adverse consequences to animal well-being (such as re-heat coils that fail closed or that are equipped with high-temperature cut-off systems), and
- how room temperature, ventilation, and critical air pressures are monitored and maintained in the event of a system or component failure, including notifying appropriate personnel in the event of a significant failure that occurs outside of regular working hours and/or other management systems used to respond to alerts or failures.

- The vivarium has a dedicated air handling unit which provides 100% fresh air, with no recirculation, to the animal facilities after treatment through a 30% efficiency pleated prefilter and 95% efficiency bag-type after filter. Housing room air is further filtered through ventilated cage racks' HEPA filtered supply air blower to individual cages.
- The air handing unit operates in variable air volume (VAV) mode, maintaining continuous air volumes of 10 or greater air exchanges per hour in all animal housing rooms. Each temperature control zone, e.g each housing room for room level control, is served with a supply VAV box with an integral reheat coil and a control valve to modulate flow of hot water to the reheat coil to maintain zone temperature conditions. Each supply VAV box is set to "track and trail" airflow of its complementary exhaust VAV box to modulate damper positions to maintain required airflow (with matching minimum-maximum cubic feet per minute set-points) as system pressures fluctuate and to maintain negative pressure gradients (or reciprocally for positive pressure gradients). Housing, testing, and procedure rooms for animals are maintained under negative pressure gradients, and personnel areas are maintained under positive pressure gradient to animal service corridors. A minimum of ~ 50 cfm volumetric offset is used to maintain room air differential pressures. Chilled water from a centralized plant serves the air handling unit's cooling coil bank and air is further tempered by re-heat coils in each room's VAV supply box. Centralized boilers generate steam to support preheat, reheat water, humidification, and domestic/laboratory hot water demand of the building. Humidification is added centrally for zonal humidification, and all housing rooms including laboratory housing room have trim

humidifiers to provide room level control. HVAC measurements are performed triennially by a commercial air balancing vendor to verify the automated building systems' performance.

- The Redacted by has two types of reheat coil control valves. The electronic reheat coil control valve, used within the vivarium, fails in the position of last command pneumatic reheat coil control valves, used in personnel spaces, fail in the closed position. Air handlers and boilers but not chillers are supported by emergency power generators; chillers retain some cooling capacity through residual chilled water in underground piping between plant and building for some time after power loss. The building automation system automatically alerts building engineers of temperature excursions. Building engineers respond by being able to adjustment HVAC operation remotely as well as on-site by rapid recruitment of first-responders.
- The building automation system continually monitors supply and exhaust air flows and temperatures of room exhaust air. Room level control of supply air flow is programmed to trail exhaust air flow to maintain critical negative differential air pressures, e.g. ABSL-2, microPET, and quarantine suites, between room and corridor, i.e. negative pressures are preserved as ventilation rates decrease towards a static, neutral pressure state. The building automation system provides telephonic and electronic notification to building engineers and animal facility managers of temperature and humidity deviations beyond programmed alarm points. Animal Facility design anticipates coupling the HVAC duct system to an exterior sited, HVAC unit to provide treated air in event of a future prolonged, catastrophic system failure.

The Animal Program's Disaster Response Plan includes coordinated responses by the animal care program staff and building engineers:

- Supply air flow can be reduced at the room level to prevent further excursion in room temperature deviation until correction of a component failure.
- Ventilation can be reduced to conserve conditioned air within a room.
- Rooms can be ventilated through partially opened doors with tempered air from animal facility corridors
- Electric, oil-filled space heaters and water-supplied spot coolers, which are stored within the animal facility, can be deployed within critical rooms. Additional heaters and coolers are readily available from local commercial sources.
- Breeding colony rodents can be relocated to the Redacted by agreement animal facility, and animals can be relocated to other areas of the Redacted by agreement with functional HVAC system components in event of prolonged system or component failure.

In the Table below, provide room-specific information requested. For each room within this location, indicate use, including the species for animal housing rooms. *Measurement of air exchange rates and verification of relative pressure within animal housing rooms (excluding rooms housing aquatic species only) and cage washing facilities must be completed* ***within the 12 months preceding completion of this Program Description.*** Air exchange rates may be important to maintain air quality in other areas; *however, measurements may be left at the discretion of the institution.* Information may be provided in another format, providing all requested data is included. **[*Note*: Please remove the examples provided in the Table below.]**

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Laboratory, support, rodent	70 F	Y	+/- 4 F from setpoint	Y	-	27.4	5/2018
	Laboratory, testing, rodent	72 F	Y	+/- 4 F	Y	-	19.8	5/2018
	Laboratory, testing, rodent	72 F	Y	+/- 4 F	Y	-	18.9	5/2018
	Laboratory, testing, rodent	72 F	Y	+/- 4 F	Y	-	10.6	5/2018
	Laboratory, testing, rodent	None, corridor controls	Y	None	Y	-	6.2	5/2018
	Laboratory, testing, rodent	68 F	Y	+/- 4 F	Y	-	19.6	5/2018
	Laboratory, testing, rodent	73 F	Y	+/- 4 F	Y	-	19.3	5/2018
	Laboratory, testing, rodent	73 F	Y	+/- 4 F	Y	-	20.4	5/2018
	Laboratory, testing, rodent	73 F	Y	+/- 4 F	Y	-	18.3	5/2018
	Laboratory, holding, rodent	72 F	Y	+/- 4 F	Y	-	18.5	5/2018
	Laboratory, surgery, rodent	73 F	Y	+/- 4 F	Y	-	20.4	5/2018
	Laboratory, office	73 F	Y	+/- 4 F	Y	-	14.5	5/2018
	MicroPET imaging	73 F	Y	+/- 4 F	Y	-	17.0	5/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Laboratory, testing, rodent	N/A, Corridor	Y	None	Y	-	6.2	5/2018
	Laboratory, procedure	72 F	Y	+/- 4 F from setpoint	Y	-	12.3	5/2018
	Laboratory, support	72 F	Y	+/- 4 F	Y	+	20.8	5/2018
	Laboratory, surgery, rodent	72 F	Y	+/- 4 F	Y	-	16.0	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	34.5	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	14.8	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	18.8	5/2018
	Laboratory, support	72 F	Y	+/- 4 F	Y	-	18.6	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	21.6	5/2018
	Laboratory, support	72 F	Y	+/- 4 F	Y	+	25.8	5/2018
	Laboratory, procedure, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	25.1	5/2018
	Laboratory, surgery, rodent	68 F	Y	+/- 4 F	Y	-	24.4	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	13.6	5/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Laboratory, necropsy, rodent	72 F	Y	+/- 4 F	Y	-	43.7	5/2018
	Laboratory, support, rodent	72 F	Y	+/- 4 F from setpoint	Y	-	20.3	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	17.4	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	17.2	5/2018
	Laboratory, surgery, rodent	72 F	Y	+/- 4 F	Y	-	15.8	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	16.2	5/2018
	Laboratory, surgery, rodent	72 F	Y	+/- 4 F	Y	-	11.2	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	15.8	5/2018
	Laboratory, support	72 F	Y	+/- 4 F	Y	-	18.8	5/2018
	Laboratory, support	72 F	Y	+/- 4 F	Y	-	18.1	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	17.4	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	17.1	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	17.0	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	17.5	5/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Laboratory, procedure, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	16.9	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F from setpoint	Y	-	17.0	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	16.6	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	16.0	5/2018
	Laboratory, procedure, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	21.3	5/2018
	Laboratory, procedure, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	19.3	5/2018
	Laboratory, procedure, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	15.5	5/2018
	Laboratory, procedure, rodent	74 F	Y	+/- 4 F	Y	-	25.4	5/2018
	Laboratory, procedure, rodent	74 F	Y	+/- 4 F	Y	-	14.6	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	+	18.4	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	18.2	5/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	10.1	5/2018
	Laboratory, procedure, rodent	69 F	Y	+/- 4 F from setpoint	Y	-	31.5	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	29.6	5/2018
	Laboratory, procedure, rodent	73 F	Y	+/- 4 F	Y	-	21.1	5/2018
	Laboratory, procedure, rodent	69 F	Y	+/- 4 F	Y	-	21.6	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	18.2	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	15.3	5/2018
	Laboratory, procedure, rodent	70 F	Y	+/- 4 F	Y	-	20.0	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	30.8	5/2018
	Laboratory, procedure, ASBL-2, rodent	68 F	Y	+/- 4 F	Y	-	13.2	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	17.1	5/2018
	Cagewash, dirty	68 F	Y	+/- 4 F	Y	-	>20.4	5/2018
	Support, storage	68 F	Y	+/- 4 F	Y	+	15.3	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	17.1	5/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	16.0	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F from setpoint	Y	-	17.7	5/2018
	Laboratory, support	72 F	Y	+/- 4 F	Y	+	17.5	5/2018
	Laboratory, support, pharmacy	72 F	Y	+/- 4 F	Y	+	14.8	5/2018
	Laboratory, support	72 F	Y	+/- 4 F	Y	+	16.5	5/2018
	Laboratory, procedure, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	17.9	5/2018
	Laboratory, procedure, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	15.9	5/2018
	Laboratory, administrative, office	73.5 F	Y	+/- 4 F	Y	+	17.1	5/2018
	Laboratory, procedure	72 F	Y	+/- 4 F	Y	-	16	5/2018
	Necropsy, shared	72 F	Y	+/- 4 F	Y	-	32	5/2018
	Quarantine receiving	72 F	Y	+/- 4 F	Y	-	18.9	5/2018
	Storage, bedding	72 F	Y	+/- 4 F	Y	-	16.6	5/2018
	Storage, general	72 F	Y	+/- 4 F	Y	-	17.5	5/2018
	Storage, food	66 F	Y	+/- 4 F	Y	-	15.6	5/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Storage, general	72 F	Y	+/- 4 F	Y	+	66.7	5/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	16.3	5/2018
	Laboratory, support, pharmacy	72 F	Y	+/- 4 F from setpoint	Y	+	17.5	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	14.8	5/2018
	Laboratory, surgery, rodent	72 F	Y	+/- 4 F	Y	+	14.8	5/2018
	Laboratory, procedure, rodent	68 F	Y	+/- 4 F	Y	-	17.3	5/2018
	Laboratory, support, Rodent	74 F	Y	+/- 4 F	Y	+	15.0	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	16.8	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	17.6	5/2018
	Laboratory, procedure, ASBL-2, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	15.5	5/2018
	Laundry	72 F	Y	+/- 4 F	Y	-	17.3	5/2018
	Laboratory, procedure, ASBL-2, rodent	68 F	Y	+/- 4 F	Y	-	20.4	5/2018
	Animal holding, quarantine	70 F	Y	+/- 4 F	Y	-	22.0	5/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Administrative, office	72 F	Y	+/- 4 F	Y	+	24.3	5/2018
	Animal holding, breeding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y		13.7	5/2018
	Administrative, office	None, corridor controls	N	None	Y	-	3.6	5/2018
	Animal holding, breeding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	15.4	5/2018
	Animal holding, breeding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	12.7	5/2018
	Administrative, break room	72 F	Y	+/- 4 F	Y	+	12.7	5/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	12.8	5/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	18.2	5/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	16.9	5/2018
	Support, water bottle fill/storage, rodent	72 F	Y	+/- 4 F	Y	+	8.3	5/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Cagewash, clean	70 F	Y	+/- 4 F	Y	- (to corridor) + (to dirty)	>33.2	5/2018
	Storage, clean caging	72 F	Y	+/- 4 F	Y	+	15.7	5/2018
	Laboratory, surgery, rodent	72 F	Y	+/- 4 F	Y	-	18.4	5/2018
	Administrative, office	70 F	Y	+/- 4 F	Y	+	9.2	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	29.1	5/2018
	Administrative, office	N/A, C626 controls	N	+/- 4 F	Y	+	8.8	5/2018
	Administrative, office	N/A, C626 controls	N	+/- 4 F	Y	+	6.0	5/2018
	Support, water bottle fill/storage, NHP	N/A, C723 controls	N	+/- 4 F	Y	+	8.9	5/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	17.2	5/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	13.8	5/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	16.0	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	22.9	5/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	16.1	5/2018
	Laboratory, procedure rodent	74 F	Y	+/- 4 F	Y	-	17.5	5/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	15.7	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	33.5	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	16.4	5/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	19.6	5/2018
	Laboratory, surgery, rodent	72 F	Y	+/- 4 F	Y	-	21.7	5/2018
	Laboratory, surgery, rodent/NHP fMRI	68 F	Y	+/- 4 F	Y	-	66.1	5/2018
	Laboratory, procedure, rodent	70 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	16.4	5/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	17.2	5/2018
	Laboratory, procedure, rodent	N/A, corridor controls	N	+/- 4 F	Y	-	8.7	5/2018
	Support, storage	72 F	Y	+/- 4 F	Y	-	17.2	5/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Laboratory, procedure, rodent	72 F	Y	+/- 4 F	Y	-	17.3	5/2018
	Animal holding, NHP	78 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	15.5	5/2018
	Laboratory, procedure	72 F	Y	+/- 4 F	Y	-	11.1	5/2018
	Animal holding, NHP	78 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	17.9	5/2018
	Laboratory, procedure, NHP	72 F	Y	+/- 4 F	Y	-	16.9	5/2018
	Animal holding, NHP	78 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	17.5	5/2018
	Laboratory, procedure, NHP	74 F	Y	+/- 4 F	Y	-	18.7	5/2018
	Support, laboratory	72 F	Y	+/- 4 F	Y	+	26.7	5/2018
	Animal holding, NHP	76 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	16.2	5/2018
	Laboratory, procedure, NHP	72 F	Y	+/- 4 F	Y	-	17.8	5/2018
	Laboratory, procedure NHP	70 F	Y	+/- 4 F	Y	-	13.4	5/2018
	Administrative, office	72 F	Y	+/- 4 F	Y	+	6.0	5/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Support, veterinary care	72 F	Y	+/- 4 F	Y	-	18.6	5/2018
	Support, surgical instrument prep	72 F	Y	+/- 4 F	Y	-	17.4	5/2018
	Surgery, NHP operating room	72 F	Y	+/- 4 F	Y	+	16.9	5/2018
	Laboratory, in vitro electrophysiology	72 F	Y	+/- 4 F	Y	+	5.2	5/2018
	Support, darkroom	N/A, corridor controls	N	+/- 4 F	Y	-	32.8	5/2018

[Create additional rows by pressing TAB in the bottom-right box.]

Copy and repeat the Description and Table for each location, including all satellite housing locations.

Location/Building/Facility:	Redacted by agreement

In the text box below, provide a general description of the mechanical systems used to provide temperature, humidity and air pressure control. Include details such as:

- the source(s) of air and air recirculation rates if other than 100% fresh air
- treatment of air (filters, absorbers, *etc.*)
- design features such as centralized chilled water, re-heat coils (steam or hot water), individual room vs. zonal temperature and relative humidity control, the use of variable air volume (VAV) systems and other key features of HVAC systems affecting performance
- features that minimize the potential for adverse consequences to animal well-being (such as re-heat coils that fail closed or that are equipped with high-temperature cut-off systems), and

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

- how room temperature, ventilation, and critical air pressures are monitored and maintained in the event of a system or component failure, including notifying appropriate personnel in the event of a significant failure that occurs outside of regular working hours and/or other management systems used to respond to alerts or failures.

- The vivarium has a dedicated air handling unit which provides 100% fresh air, with no recirculation, to the animal facilities after treatment through a 30% efficiency pleated prefilter and 95% efficiency bag-type after filter. Housing room air is further filtered through ventilated cage racks' HEPA filtered supply air blower to individual cages. Air is removed from housing rooms through a 10% efficiency, disposable fiberglass media filter at the individual room level to the exterior at roof level.

- The air handing unit operates as constant volume (CV) system, maintaining static air pressure to control CFM to provide 10 or greater air exchanges per hour in all animal housing rooms. Each temperature control zone, e.g each housing room for room level control, is served with a supply CV box with an integral reheat coil and a control valve to modulate flow of hot water to the reheat coil to maintain zone temperature conditions. Housing, testing, and procedure rooms for animals are maintained under negative pressure gradients, and personnel areas are maintained under positive pressure gradient to animal service corridors. A minimum of ~ 50 cfm volumetric offset is used to maintain room air differential pressures. Chilled water from a centralized plant serves the air handling unit's cooling coil bank and air is further tempered by re-heat coils in each room's CV supply box. Centralized boilers generate steam to support preheat, reheat water, humidification, and domestic/laboratory hot water demand of the building. Humidification is added centrally for zonal humidification, and all housing and vivarium testing rooms have trim humidifiers to provide room level control. HVAC measurements are performed triennially by a commercial air balancing vendor to verify the automated building systems' performance.

- The [Redacted by agreement] uses electronic reheat control valves which fail in the position of last command. Redundant temperature sensors are used in housing room to anticipate failure of a single sensor, and alarms activate from temperature excursion detected by either sensor. Air handlers, chillers, and boilers are supported by emergency power, a redundant air handler for the vivarium is accessible by manual damper adjustment. The building automation system automatically alerts building engineers of temperature excursions. Building engineers respond by being able to adjustment HVAC operation remotely as well as on-site by rapid recruitment of first-responders.

- The building automation system continually monitors supply and exhaust air flows and temperatures of room exhaust air. The building automation system provides telephonic and electronic notification to building engineers and animal facility managers of temperature and humidity deviations beyond programmed alarm points. Building engineers respond on-site by rapid recruitment of first-responders. If the vivarium's air handler fails, [Redacted by agreement] design provides access through a manually operated damper to access ventilation from another air handler.

The Animal Program's Disaster Response Plan includes coordinated responses by the animal care program staff and building engineers, as described in the [Redacted] summary section, above.

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
Redacted by agreement	Administrative, break room	72 F	Y	+/- 3 F from set point	Y	+	9.5	7/2018
	Administrative, office	72 F	Y	+/- 3 F	Y	+	10.4	7/2018
	Administrative, office	72 F	Y	+/- 3 F	Y	+	8.6	7/2018
	Administrative, locker room	72 F	Y	+/- 3 F	Y	-	6.9	7/2018
	Administrative, locker room	72 F	Y	+/- 3 F	Y	-	8.4	7/2018
	Storage, general	72 F	Y	+/- 3 F	Y	-	18.2	7/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 3 F	Y	-	23.1	7/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 3 F	Y	-	18.7	7/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 3 F	Y	-	20.1	7/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 3 F	Y	-	26.7	7/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 3 F	Y	-	26.1	7/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 3 F	Y	-	26.4	7/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 3 F	Y	-	12.2	7/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 3 F	Y	-	25.7	7/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

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		(settings to be verified)					(values to be measured)	
Redacted by agreement	Laboratory, procedure, rodent	72 F	Y	+/- 3 F	Y	-	14.5	7/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 3 F	Y	-	9.8	7/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	14.9	7/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 3 F	Y	-	22.6	7/2018
	Laboratory, procedure, rodent	72 F	Y	+/- 3 F	Y	-	18.9	7/2018
	Cagewash, dirty	70 F	Y	+/- 3 F	Y	+	42.8	7/2018
	Cagewash, clean; storage-caging	71 F	Y	+/- 3 F	Y	+	16.8	7/2018
	Necropsy, shared	72 F	Y	+/- 3 F	Y	-	28	7/2018
	Procedure, surgery, rodent	72 F	Y	+/- 3 F	Y	-	13.5	7/2018
	Storage, bedding & feed	66 F	Y	+/- 3 F	Y	-	14.7	7/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	16.5	7/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	11.4	7/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert)	Y	-	16.5	7/2018

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

Room No.	Specific Use	Temperature Set-Point (define units)	Electronic / Emergency Monitoring of Temperatures (Y/N)	Alert/Alarm Temperature Ranges (if applicable; define units)	Humidity Control (Y/N)	Relative Pressure	Air Exchange Rate (per hour)	Date Verified / Measured
		(settings to be verified)					(values to be measured)	
				64-78°F (critical alarm)				
Redacted by agreement	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	10.8	7/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm))	Y	-	15.4	7/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	15.3	7/2018
	Animal holding, rodent	72 F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	13.4	7/2018

Location/Building/Facility:	Redacted by agreement	

In the text box below, provide a general description of the mechanical systems used to provide temperature, humidity and air pressure control. Include details such as:

- the source(s) of air and air recirculation rates if other than 100% fresh air
- treatment of air (filters, absorbers, *etc.*)
- design features such as centralized chilled water, re-heat coils (steam or hot water), individual room vs. zonal temperature and relative humidity control, the use of variable air volume (VAV) systems and other key features of HVAC systems affecting performance

Appendix 11: Heating, Ventilation and Air Conditioning (HVAC) System Summary

- features that minimize the potential for adverse consequences to animal well-being (such as re-heat coils that fail closed or that are equipped with high-temperature cut-off systems), and
- how room temperature, ventilation, and critical air pressures are monitored and maintained in the event of a system or component failure, including notifying appropriate personnel in the event of a significant failure that occurs outside of regular working hours and/or other management systems used to respond to alerts or failures.

Please refer to the general description of the mechanical systems used to provide temperature, humidity and air pressure control for the [Redacted by agreement] above.

A wall mounted, Armstrong EHU-700 humidifier provides room-level, supplementary humidification to zonal laboratory humidity levels in mouse satellite housing rooms [Redacted by agreement]

Room No.	**Specific Use**	**Temperature Set-Point** (define units)	**Electronic / Emergency Monitoring of Temperatures** (Y/N)	**Alert/Alarm Temperature Ranges** (if applicable; define units)	**Humidity Control** (Y/N)	**Relative Pressure**	**Air Exchange Rate** (per hour)	**Date Verified / Measured**
		(settings to be verified)					**(values to be measured)**	
Redacted by agreement	**Laboratory, holding/procedure, mice**	72°F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	15.1	7/2018
	Laboratory, holding/procedure, mice	72°F	Y	69-75°F (alert) 64-78°F (critical alarm)	Y	-	25.1	7/2018

Appendix 12: Aquatic Systems Summary – Part I

Please summarize water management and monitoring information programs for each animal facility, including all satellite facilities, rooms, enclosures. The following key will assist you in completing the form:

(1) List location of aquaria, including outdoor enclosures (ponds or outdoor tanks). If indoors, list building and room number. Note that all species housed at the same location and maintained via the same design and monitoring may be listed in the same row.
(2) Please indicate if embryonic (E), larval (L), juvenile (J) or Adult (A)
(3) Group tanks (ponds, outdoor tanks, multiple aquaria) are arranged as arrays with shared water supply; individual aquaria have exclusive water handling systems.
(4) Indicate water type, e.g., fresh, brackish, or marine.
(5) Indicate water pre-treatment, e.g., dechlorination, rough filters.
(6) Indicate water circulation, e.g., static, re-circulated, constant flow, or some combination of these. If applicable, indicate water exchange frequency and amount (percentage).
(7) Provide a key word for filtration employed, e.g., biological, chemical, mechanical, and type (e.g., mechanical-bead filter). A diagram may be provided showing the flow of water, filtration, source of "make-up" water and amount replaced daily.

Part I

Location (1)	Species (2)	System Design					
		Group / Individual (3)	Water Type (4)	Pre-treatment (5)	Circulation (6)	Filtration (7)	Disinfection (e.g., UV, ozone)
NOT APPLICABLE							

Note: Records of equipment maintenance (filter changes, UV bulb changes, probe changes, calibrations, *etc.*) should be available for review.

[Create additional rows by pressing TAB in the bottom-right box.]

Appendix 12: Aquatic Systems Summary – Part II

The following key will assist you in completing this form:

(1) In these columns, please indicate monitoring frequency, e.g. daily, weekly, monthly or other point sampling frequency; continuous/real time, or none, if applicable. Also indicate method of control (heaters versus room HVAC, hand versus auto dosing, etc.).

(2) Indicate other parameters and their monitoring frequency, e.g., alkalinity, total hardness, conductivity, chlorine/chloramine.

Part II

Monitoring ***Indicate in the boxes below the frequency of monitoring and method of control for the following parameters. (1)***									
Location (from Part I)	**Temperature**	**Salinity**	**pH**	**NH_4**	**NO_2**	**NO_3**	**Dissolved O_2**	**Total Dissolved Gases**	**Other. Please List (2):**
NOT APPLICABLE									

Note: This information may be provided in another format, provided that all requested data is included.

[Create additional rows by pressing TAB in the bottom-right box.]

Appendix 13: Primary Enclosures and Animal Space Provisions

Please complete the Table below considering performance criteria and guiding documents (e.g., Guide, Ag Guide, ETS 123 and/or other applicable standards) used by the IACUC/OB to establish adequacy of space provided for all research animals including traditional laboratory species, agricultural animals, aquatic species, and wildlife when reviewing biomedical, field, and agricultural research studies.

Species	Dimensions of Enclosure (cage, pen, tank*, corral, paddock, etc.)	Maximum Number Animals / Enclosure	Guiding Document	Enclosure Composition & Description**
Mouse	11.75" L x 6.375" W x 5.625"H (IVCS, Super Mouse 750™; 74.9 in^2 flooring)	5 adults; 1 female with litter + 1 adult	*The Guide for the Care and Use of Laboratory Animals*	Mice and rats are centrally housed in polycarbonate shoebox style caging with microisolator lid covers. Operant conditioning chambers can serve as the primary enclosure during experimental sessions for behavioral testing and satellite housing; see section B.1.a.2.b., immediately below, for description of ventilated enclosures. Commercially-obtained operant conditioning chambers are constructed of stainless steel or aluminum and Plexiglas® or the equivalent. The front and back walls of the chambers are metal and the sides are transparent plastic. The floor is constructed of a series of metal rods that allow urine or feces to pass to a metal or plastic excreta tray underneath; absorbent bedding or a paper liner is contained in the tray. A reward pellet dispenser provides food pellets or a liquid reward dispenser provides palatable solutions during sessions if required by the experimental protocol.
Mouse	10.75" L x 6.375" W x 5.0" H (Static, conventional; 68.53 in^2 flooring)	4 adults; 1 female with litter + 1 adult		
Mouse	6.5" L x 5.5" W x 5" H (Operant chamber; 35.75in^2 flooring)	1 adult		
Rat	12.25"L x 7.25" W x 7.375" H (IVCS, OneCage ™; 88.8 in2 floor)	2 adults; Not to exceed 750 gm Cumulative BW		
Rat	13.75"Lx11.0"Wx7.375"H (IVCS, Super Rat 1400 ™ ; 151.25 in2 floor)	4 adults, Not to exceed 1000 gm cumulative BW; or 1 female with litter		
Rat	12.25"Lx17.25"Wx7.5"H (IVCS, OneCage 2100 ™; 210 in^2 floor)	2 adults with one litter, not to exceed 1500 gm cumulative BW		

Appendix 13: Primary Enclosures and Animal Space Provisions

Species	Dimensions of Enclosure (cage, pen, tank*, corral, paddock, etc.)	Maximum Number Animals / Enclosure	Guiding Document	Enclosure Composition & Description**
Rat	16.5" L x 8.5" W x 8" H (Static, conventional; 140.25 in^2 flooring)	3 adults; 1000 gm maximum total BW		
Rat	11.5" L x 9.5" W x 8" H (Operant chamber; 109.2 in^2 flooring)	1 adult		
Squirrel monkey	27"Lx30"Hx26"H (4-compartments, 5.6 ft^2 floor)	1-1.5; 3 per tier		Nonhuman primates (NHP) are housed in conventional stainless steel caging which can be reconfigured to support social housing within the rack or between docked racks.. • The four compartment system has fixed grid floors with a removable common wall panel. • The six compartment system has removable flat bar floors and repositionable common walls. • The two-compartment system has retractable grid floors and common walls. and two compartment systems
Squirrel monkey	27"Lx19"Wx30.5"H (6-compartments, 3.56 ft^2 floor)	1-1.5; 3 per tier		
Rhesus monkey	39"Lx33"Wx 33"H, (2-compartment, 8.9 ft^2 floor)	1 adult		

*For aquatic species, provide tank volume.

**Include descriptors such as open-topped, static microisolator, individually-ventilated cage systems (IVCS).

Appendix 14: Cleaning and Disinfection of the Micro- and Macro-Environment

Please describe the cleaning and disinfection methods in the Table below. Note the washing/sanitizing frequency and method for each of the following:

Area	Washing/Sanitizing Method (mechanical washer, hand washing, high-pressure sprayers, etc.)	Washing/ Sanitizing Frequency	Chemical(s) Used*	Other Comments (e.g., autoclaved)
Micro-environment				
Solid-bottom cages (static)	Mechanical washer	Weekly – individually housed Biweekly (two times a week) – group housed	Labsan 120 (Sanitation Strategies, Holt, MI) is a proprietary mixture of detergents in a sodium metasilicate alkalinized solution with Ethylenediaminetetraacetic Acid (EDTA) for chelation and other water softeners.	
Solid-bottom cages (IVC)	Mechanical washer	Weekly	Labsan 120	
Suspended wire-bottom or slotted floor cages	Hand washing for pre-wash processing. Mechanical washer for sanitizing	Weekly	Chlorfoam Plus 120 (Sanitation Strategies, Holt, MI) is a chlorinated (Sodium hypochlorite), alkalinized detergent solution with caustics (potassium hydroxide and sodium phosphate) and proprietary foaming agents for pre-wash processing. Labsan 120 (Mechanical washing)	NHP cage racks is being described here. See "Rodent operant chambers" regarding their slotted, metal rod floors.
Cage lids	Mechanical washer	Every two weeks	Labsan 120	

Appendix 14: Cleaning and Disinfection of the Micro- and Macro-Environment

Area	Washing/Sanitizing Method (mechanical washer, hand washing, high-pressure sprayers, etc.)	Washing/ Sanitizing Frequency	Chemical(s) Used*	Other Comments (e.g., autoclaved)
Filter tops	Mechanical washer	Every two weeks	Labsan 120	Filter tops are integrated into cage lids.
Cage racks and shelves	Mechanical washer	Semiannually (IVC)	Labsan 120	
Cage pans under suspended cages		Three times a week	Labsan 230C (Sanitation Strategies, Holt, MI) is 30% citric acid based detergent solution with a a proprietary mixture of surfactants.	NHP excreta pans. See "Rodent operant chambers" regarding their excreta pans under slotted floors.
Play pens, floor pens, stalls, etc.				N/A
Corrals for primates or outdoor paddocks for livestock				N/A
Aquatic, amphibian, and reptile tanks and enclosures				N/A
Feeders	Mechanical washer	Weekly (NHP) Every two weeks (Rodents)	Labsan 120	
Watering devices	Mechanical washer	Weekly (Rodent Bottles) Biweekly (NHP bottles)	Labsan 120	NHP water manifolds are

Appendix 14: Cleaning and Disinfection of the Micro- and Macro-Environment

Area	Washing/Sanitizing Method (mechanical washer, hand washing, high-pressure sprayers, etc.)	Washing/ Sanitizing Frequency	Chemical(s) Used*	Other Comments (e.g., autoclaved)
				sanitized with cage.
Exercise devices and manipulanda used in environmental enrichment programs, etc.	Mechanical washer	Weekly	Labsan 120	NHP manipulanda sanitized along with cage.
Transport cages	Mechanical washer	Daily (NHP)	Labsan 120	A disposable paper lining is replaced between Squirrel monkey occupants for transporting between housing and testing rooms.
Operant conditioning & recording chambers, mechanical restraint devices (chairs, slings, etc.)	Hand washing	Weekly (NHP chairs) Every two weeks (Rodent operant chamber)	Quaternary ammonia (e.g. Labsan 256CPQ) Chlorine dioxide solution Alcohols (70% ethanol, isopropanol) Dishwashing detergent	
Euthanasia chambers	Hand washing	Daily, after use	Labsan 256CPQ (Sanitation Strategies, Holt, MI) is a mixture of quaternary ammonium compounds	CO2 lid used upon

Appendix 14: Cleaning and Disinfection of the Micro- and Macro-Environment

Area	Washing/Sanitizing Method (mechanical washer, hand washing, high-pressure sprayers, etc.)	Washing/ Sanitizing Frequency	Chemical(s) Used*	Other Comments (e.g., autoclaved)
			(Dideclydimethylammonium Chloride, Octyldimethyl Amine Oxide and Alkyl Dimethyl Benzyl Ammonium Chloride) and Ethylenediaminetetraacetic Acid (EDTA), Tetrasodium Salt for chelation.	polycarbonate cage.
Macro-Environment				
Animal Housing Rooms:				
Floors	Hand washing	Daily	NHP areas: Quatricide TB (Pharmacal Research Laboratories, Naugatuck, CT) is an mixture of quaternary ammonia compounds (n-Alkyl dimethyl benzyl ammonium chlorides, n-Alkyl dimethyl ethylbenzyl ammonium chlorides) Labsan 256 (Rodent)	
Walls	Foam and water rinse applicator, wet vacuum removal	Monthly (NHP) Semiannual (Rodent)		
Ceilings		Monthly (NHP) Semiannual (Rodent)		
Ducts/Pipes		Monthly (NHP) Semiannual (Rodent)		
Fixtures		Monthly (NHP) Semiannual (Rodent)		

Appendix 14: Cleaning and Disinfection of the Micro- and Macro-Environment

Area	Washing/Sanitizing Method (mechanical washer, hand washing, high-pressure sprayers, etc.)	Washing/ Sanitizing Frequency	Chemical(s) Used*	Other Comments (e.g., autoclaved)
Corridors:				
Floors	Mechanical [Redacted by] Hand washing [Redacted by agreement]	Weekly [Redacted by] Daily [Redacted by agreement]	Quatricide TB (NHP) Labsan 256 (Rodent)	
Walls	Foam and water rinse applicator, wet vacuum removal	Monthly		
Ceilings				
Ducts/Pipes				
Fixtures				
Support Areas (e.g., surgery, procedure rooms, etc.); complete for each area:				
Floors	Hand washing and Mechanical [Redacted by] Hand washing (Triad)	Daily (hand) and quarterly (mechanical) - [Redacted] surgery suite Semiannually (mechanical) – all other support areas (e.g. storage rooms, cagewash area)	Labsan 256 CPQ	
Walls	Foam and water rinse applicator, wet vacuum removal			
Ceilings				
Ducts/Pipes				
Fixtures				
Implements (note whether or not shared): not shared				

Appendix 14: Cleaning and Disinfection of the Micro- and Macro-Environment

Area	Washing/Sanitizing Method (mechanical washer, hand washing, high-pressure sprayers, etc.)	Washing/ Sanitizing Frequency	Chemical(s) Used*	Other Comments (e.g., autoclaved)
Mops	Mechanical washer	Weekly	Labsan 120	
Mop buckets	Mechanical washer	Weekly		
Aquaria nets				N/A
Other				
Other:				
Vehicle(s)	Hand wash	Quarterly	Labsan 256 CPQ	Dedicated animal transport van
Other transport equipment (list)				

*Please provide chemical, not trade name.

Appendix 15: Facilities and Equipment for Sanitizing Materials

In the Tables below, summarize the facilities and equipment used to sanitize animal related equipment (tunnel washer, bottle washer, rack washer, bulk autoclave, hand-washing area, bedding dispensing unit, *etc.*). Note that some descriptions may be combined if all share identical features (e.g., all rack washers).

[*Note*: Please remove the examples provided in the Table below.]

Building	Room No.	Equipment Type	Safety Feature(s)	Methods of Monitoring Effectiveness
Redacted by agreement	Redacted by agreement	Rack washer	Emergency "off" button; de-energizing cord on both sides; labeled exit door, instructional signs, and explosion relief door latches on both doors.	Mechanical washers: guarantee 180-degree hot water rinse; temperature-sensitive tape used daily; visual assessment; ATP-based luminescence swabs performed monthly.
		Tunnel Washer	Emergency "off" button; de-energizing cord on both ends	
		Bedding dispensing unit	Emergency "off" button	
Redacted by agreement	Redacted by agreement	Rack washer	Emergency "off" button; de-energizing cord on both sides; labeled exit door, instructional signs, and explosion relief door latches on both doors	
		Tunnel Washer	Emergency "off" button; Emergency "off" button; de-energizing cord on both ends	
		Bedding dispensing unit	Emergency "off" button	

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Appendix 16: Lighting Summary

Using the Table below, summarize the lighting system(s) for the animal housing facility(ies). For each species or holding room type, list light intensity (range), construction features (e.g., water resistance), photoperiod (light:dark) and control (e.g., automatic versus manual, phasing). For systems automatically controlling photoperiod, describe override mechanisms (including alarms, if applicable).

Location: Redacted by agreement

[*Note*: Please remove the examples provided in the Table below.]

Room Type[a]	**Light Intensity Range**	**Lighting Fixture Construction Features**[b]	**Photo-period** (hrs)[c]	**Photoperiod and Lighting Control**	**Override Mechanisms** (if applicable)
Animal holding, rodent	25-30 Foot-candles (Fc) or 45-50 Fc	• Recessed, water resistant ceiling fixtures with white fluorescent lamps and with red color, plastic sleeved fluorescent lamps • Ceiling mounted, red globed jelly jar style fixtures with incandescent or compact fluorescent lamps.	12:12 Five of 13 rooms operate under reversed light cycles.	Automatic periodicity lighting is under control of the Building Automation System (BAS control).	Wall switches allow activation of: • White fluorescent lamps to activate dual level light levels only during light phase of standard cycle. • Red sleeved fluorescent lamps and, via separate wall switch, an electronic timer, activates red globed jelly jar fixtures for 3 hours.
Animal holding, NHP	45-50 Fc or 75-80 Fc	• Recessed, water resistant ceiling fixtures with white fluorescent lamps	12:12 Standard Light Cycle	BAS control	Wall switches allow activation of white fluorescent lamps to activate dual level light levels only during light phase of cycle.

Appendix 16: Lighting Summary

Room Type[a]	**Light Intensity Range**	**Lighting Fixture Construction Features**[b]	**Photo-period** (hrs)[c]	**Photoperiod and Lighting Control**	**Override Mechanisms** (if applicable)
Laboratory, holding/procedure, rodents	50-80 Fc	• Recessed, water resistant ceiling fixtures with white fluorescent lamps • Ceiling mounted, red globed jelly jar style fixtures with incandescent or compact fluorescent lamps. • Sound attenuating chamber doors contain and operant chamber doors are transparent plastic windows. • Lights mounted on the top of the operant testing chamber's front wall provide additional illumination within in the chamber.	12:12 Ten of 15 laboratory holding rooms operate under reversed light cycles corresponding to animals' originating housing rooms' cycles.	BAS control	Wall switches allow activation of: • Red sleeved lamps and red globed jelly jar fixtures via electronic timer, set at 3 hours.
Animal holding, quarantine rodents, and Laboratory holding, ASBL-2 rodents.	15-30 Fc	• Recessed, water resistant ceiling fixtures with white fluorescent lamps are adjacent to the clear glass doors of the galley of cubicles. • Two vertically mounted, water resistant wall fixtures with white fluorescent lamps within each cubicle.		BAS control for the cubicle room; the wall mounted light fixtures are programmed for independent control of each cubicle.	• Wall mounted fixtures can be manually activated with a switch for each cubicle.

Appendix 16: Lighting Summary

Room Type[a]	**Light Intensity Range**	**Lighting Fixture Construction Features**[b]	**Photo-period** (hrs)[c]	**Photoperiod and Lighting Control**	**Override Mechanisms** (if applicable)
Non-rodent Surgery	50-60 Fc	• Recessed, water resistant ceiling fixtures with white fluorescent lamps. • Ceiling mounted, LED surgical lights on adjustable armature.	NA	N/A	N/A
Necropsy	50-70 Fc	• Recessed, water resistant ceiling fixtures with white fluorescent lamps.	NA	N/A	N/A
Cage-Washing Room	50-60 Fc	Recessed, water proof	NA	N/A	N/A
fMRI laboratory housing Redacted by agreement Redacted by agreement	~ 120 Fc	• Recessed, water resistant ceiling fixtures with white LED lamps.	12:12	BAS control	Electrical on/off switch
Support areas	40-80 Fc				

[Create additional rows by pressing TAB in the bottom-right box.]

Location: Redacted by agreement

Appendix 16: Lighting Summary

Room Type[a]	**Light Intensity Range**	**Lighting Fixture Construction Features**[b]	**Photo-period** (hrs)[c]	**Photoperiod and Lighting Control**	**Override Mechanisms** (if applicable)
Animal holding, rodent	25-30 Foot candles (Fc)	• Recessed, water resistant ceiling fixtures with white fluorescent lamps and with a red plastic sleeved fluorescent lamp.	12:12 One of 7 rooms operate under reversed light cycles.	Periodicity lighting is under control of the Building Automation System (BAS).	Wall switch manually activates the red sleeved fluorescent lamp in each ceiling fixture.
Laboratory, procedure, rodent	30-45 Fc	• Recessed, water resistant ceiling fixtures with white fluorescent lamps	N/A	Manual wall switch	N/A
Necropsy	30-35 Fc	• Recessed, water resistant ceiling fixtures with white fluorescent lamps	N/A	Manual wall switch	N/A
Support areas	40-45 Fc	• Recessed, water resistant ceiling fixtures with white fluorescent lamps	N/A	Manual wall switch	N/A
Cage-Washing Rooms	35-45 Fc	• Recessed, water resistant ceiling fixtures with white fluorescent lamps	N/A	Manual wall switch	N/A
Personnel areas	30-40 Fc	• Recessed, water resistant ceiling fixtures with white fluorescent lamps	N/A	Manual wall switch	N/A

Location: Redacted by agreement

Appendix 16: Lighting Summary

Room Type[a]	**Light Intensity Range**	**Lighting Fixture Construction Features**[b]	**Photo-period** (hrs)[c]	**Photoperiod and Lighting Control**	**Override Mechanisms** (if applicable)
Satellite Rodent Laboratory Housing	20-25 Fc	• Recessed, water resistant ceiling fixtures with white fluorescent lamps and with a red plastic sleeved fluorescent lamp.	12:12 Reversed light cycles	Automated periodicity lighting is under control of wall mount, digital timers for each room.	Wall mounted timer manually activates the red sleeved fluorescent lamp in each ceiling fixture.

[a] A list of each room is not needed; group or cluster rooms by species or function
[b] Include such features as water resistance, red lighting, *etc.*
[c] Note if light cycle inverted/reversed.

Repeat Location and Table as necessary for each location, including satellite housing locations.

Appendix 17: Satellite Housing Facilities

Note: In the Program Description Section 2. IV. (Physical Plant), item C., describe the criteria used to determine a "Satellite Animal Holding Area." In the Table below, summarize these animal housing areas. Note that the total square footage for all each of these must also be included in the Summary of Animal Housing and Support Sites (**Appendix 2**), and applicable information regarding these areas included in the Heating, Ventilation, and Air Conditioning (HVAC) Summary (**Appendix 11**) and Lighting Systems Summary (**Appendix 16**).

Building	Room(s)	Person Responsible	Species Used	Approximate Area (ft^2 or m^2) Devoted to Housing	Maximum Period of Stay	Purpose / Rationale / Justification	Construction Features and Finishes
Redacted by agreement	Redacted by agreement	Redacted by agreement	Mice	153 ft^2	Two months	Satellite housing avoids daily transport of mice for distances through the building for sensitive behavioral procedures.	• Epoxy paint finish over gypsum panels and metal frame walls. • Suspended, scrubable 2'x4' vinyl-faced ceiling panel ceiling. • Seamless sheet vinyl floors • Sealed wooden doors and galvanized steel frames.
Redacted by agreement	Redacted by agreement	Redacted by agreement	Mice	144 ft^2			

[Create additional rows by pressing TAB in the bottom-right box.]