



View xForm - IACUC Animal Use Protocol Form

IACUC Animal Use Protocol Form v2.1

Animal Use Protocol Form Data Entry

A. Initial Demographics

** you can use the drop-down list above to temporarily skip past error messages. *

Date for 180 day notice

10/6/2020

Submitter

Joseph, Elaine

Email: Elaine.Joseph@UToledo.Edu

Business: 4193834251

A.1. What is the title of your project?

Title should reflect content and subject of animal project

A.2. Please indicate the protocol type for your application

No answer provided.

A.3. Please indicate species involved in your protocol (one per protocol).

Entered: 10/06/20 **By:** Joseph, Elaine **Internal:** No

Only one species is allowed per protocol. If your species (or type of species) is not available, please contact IACUC@utoledo.edu

No answer provided.

A.4. Is this a renewal application?

Entered: 11/04/20 **By:** Joseph, Elaine **Internal:** No

To enter the number for protocols from the old system, please look at the study (protocol) page in IRB Manager and type the six digit number followed by the two digit number (i.e., 123456-12). For protocols created in IRB Manager, simply type the six digit number (i.e., 400123).

Yes

What protocol is this a renewal for?

No answer provided.

Note, you must type the entire number into this space. The format for numbers coming from the old system is 1#####-##. The format for numbers started in this system is 4#####. You can copy and paste from the Study screen.

A.4.2. Type of renewal

No answer provided.

A.4.3. Provide a brief summary of what has been accomplished on this protocol to date, including number of animals initially approved, number actually used, and specific aims accomplished.

This should be a brief summary of what you have accomplished (it does not need to include publications), such as experiments completed and experiments in progress. You must also include the number of animals previously approved for and the number of animals used. The number of animals used can be obtained from DLAR.

Species Contains USDA

False

Calculate Title for Review Details

Title should reflect content and subject of animal project

B. Protocol Personnel

** you can use the drop-down list above to temporarily skip past error messages. **

B.1. Select Principal Investigator by typing their last name into the box below and selecting the appropriate entry from the list.

No answer provided.

B.2. P.I. Department

No answer provided.

If the P.I.'s department does not appear, it may be because it was either missing or incorrect in the KC or RSP system or the person was created using the automatic UTAD process, which does not have access to the person's department. A connection to update this data from Banner is in progress but will not be finished for a while.

B.3. If the P.I.'s department is missing above, please choose the department here

No answer provided.

Renewal Protocol - B.4. Study-Site Contacts Instructions.

Personnel may have changed since the original Protocol xForm was approved. Please update the list with the correct protocol personnel for renewal protocol.

- 1) For anyone no longer on the protocol, use the red x at the far left of the Action column to delete that person. You will need to confirm each deletion.
- 2) For anyone still on the protocol, review the training that was requested during the original protocol or latest renewal. Most people who needed training will have been trained and thus the training columns can be blanked out. Click the "Edit" icon in the left action column, make the changes to the row, and then click the green check mark to save your changes to that row.
- 3) Add any personnel who are missing and select any needed training. You can see the current list of the protocol personnel on the main page of the expiring study from which you copied this application.

B.4. Study-Site Contacts

Entered: 10/06/20 **By:** Joseph, Elaine **Internal:** No

Even for a renewal protocol, you must put in all personnel working with animals. If they are to be an emergency contact for animal care, please send their emergency phone number to IACUC@utoledo.edu. If you, as the PI, will be working directly with the animals, please list yourself in this section.

Personnel Name	Emergency Contact?	Performing Surgical Procedures?	Needs Training?	Select the Training Needed (^ = Required for all personnel)	Enter specific technical training (e.g. Oral Gauvage, Tail Vein Injection, RO Injection)
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If you are unable to locate a UT-affiliated person in the drop down list to add to your study, ask the person to log into IRB Manager using their UTAD credentials. Within 5 minutes the person's contact record will available to add to the personnel table above.

C. Associated Protocol Information

** you can use the drop-down list above to temporarily skip past error messages. **

C.1. Will tissues from these animals be used in any recombinant DNA experiments or agents derived from recombinant DNA be introduced into these animals? Will infectious agents (virus, bacteria, etc.) be introduced into these animals? Will biohazardous agents (human tissues/fluids, transfected cells, etc.) be introduced in these animals?

Entered: 10/06/20 **By:** Joseph, Elaine **Internal:** No

The ABSL number can be obtained from the IBC that is associated with the IACUC you are proposing.

Yes

Recombinant DNA is a molecule of DNA that has been modified, either through genetic recombination or through laboratory techniques

C.1.1. Recombinant DNA-derived agents will be introduced into animals?

No answer provided.

C.1.2. Biohazardous/infectious agents will be administered to animals?

No answer provided.

C.1.3. Please select the Biosafety Level the animals should be housed (ABSL1, 2, 3)

No answer provided.

ABSL1 is appropriate for research conducted with well-characterized strains of viable microorganisms or viruses not known to cause disease in healthy adult humans.

ABSL2 refers to practices and procedures required to conduct research with animals infected with agents that can cause disease in humans. ABSL2 is designed to prevent exposures to infectious material via percutaneous injury, mucous membrane splashes and accidental ingestion.

ABSL3 involves practices suitable for work with laboratory animals infected with indigenous or exotic agents, agents that present a potential for aerosol transmission and agents causing serious or potentially lethal disease. ABSL3 builds upon the standard practices, procedures, containment equipment, and facility requirements of ABSL2.

C.1.4. Please list all relevant IBC protocol numbers:

No answer provided.

C.2. Does this protocol involve the use of any radioactive isotopes in live vertebrate animal subject?

No answer provided.

Radio isotopes are unstable isotopes used in the treatment of cancer or as tracers for diagnostic purposes, examples include cobalt-60, iodine-131, carbon-14, carbon-11, thallium-201

C.3. Does this protocol involve the use of any chemicals on or in animals (i.e., topically, injected, ingested, etc.) apart from anesthetics and analgesics?

Entered: 10/06/20 **By:** Joseph, Elaine **Internal:** No

This sections refers to any experimental or pharmaceutical chemicals that you are administering to animals as a part of the experiment. It does not include anesthetics or analgesics. For any experimental or pharmaceutical chemicals that you are administering, you must upload a chemical addendum (RSP 402) and an SDS or MSDS. The RSP 402 can be found at <https://www.utoledo.edu/research/rsp/RC/iacuc-forms.html>

Yes

Chemicals include any biological or chemical agents used in or in animals, including drugs, cell lines, etc.

C.3.1. You must submit electronic copies of the both the RSP402 (protocol addendum for use of chemicals) and the Safety Datasheet (SDS) for EACH agent along with this protocol. The protocol addendum form can be found here.

C.3.2. Please attach appropriate chemical addendum and Safety Data Sheets.
No answer provided.

C.4. Does this protocol involve field work?

No answer provided.

C.5. Is protocol supported by extramural (outside), peer-reviewed funding (e.g., NIH, NSF)?

No answer provided.

C.6. Are you housing animals as part of this protocol for more than 12 hours?

No answer provided.

D. Lay Description

** you can use the drop-down list above to temporarily skip past error messages. **

D.1. The purpose of this summary is to provide the general public a concise summary of this project. Avoid technical terms, abbreviations, and scientific jargon about the proposed experiments in this section. Remember the audience of this summary is the general public and should be aimed at an 8th grade reading level. Your summary should concisely summarize three topics:

- 1) the objectives of this study;**
- 2) why it is important; and**
- 3) why you need animals to complete this study.**

A proper lay description will avoid complicated scientific terms or will define those terms using lay language. It will also clearly state the objectives or goals of the research. You should state why the research is important in terms of either animal health, human health, or improving scientific knowledge. Finally, you should state why you need animals to complete the study, as opposed to cell lines or computer models.

E. Statistical Design and Animal Numbers

** you can use the drop-down list above to temporarily skip past error messages. **

E.1. The number of animals requested in this protocol is based on the following (select all that apply):

No answer provided.

E.2. Provide equations for the number of animals in each proposed experiment, e.g. '2 strains x 10 animals/sample x 3 drugs x 6 doses x 3 time points = 1080 animals'. Also, indicate the total number of aims/groups below. It is recommended, but not required, that you identify the strains you will use in order to facilitate the review of your experimental design and animal numbers. For breeding protocols, indicate total numbers (breeders and pups) to maintain breeding colony and associated experimental protocols. Do not forget unwanted genotypes.

This section should provide equations for each experiment or section of proposed project. For example:

Experiment 1 (determine dosage):

3 strains [inserting names of strains can be helpful] X 10 animals/sample X 3 drugs [inserting names of drugs can be helpful] X 2 doses [inserting dosages can be helpful] X 2 sexes X 2 replicates = 900

Experiment 2 (with one dose):

3 strains [inserting names of strains can be helpful] X 10 animals/sample X 3 drugs [inserting names of drugs can be helpful] X 2 sexes X 2 replicates = 360

Breeding:

4 breeder pairs (2 mice) x 6 pups per litter x 4 litters per year x 3 years = 576

E.3. Strains are not required if there is a phenotype that may impact the health of the animals. Only list those strains and the impact on their health. Do not list strains with normal phenotypes.

Entered: 11/12/20 **By:** Joseph, Elaine **Internal:** No

Please be sure to use the proper nomenclature for strains.

Strain	Phenotype(s) that May Impact Health (i.e., diabetes, immunocompromised, runted, etc.) and how you will monitor animals/address condition
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E.4. Are you going to be utilizing any special husbandry procedures as part of this protocol (i.e., special diet, special water, special bedding requirements, Specialized caging, etc.)?

No answer provided.

E.5. Enter the total number of animals to be used in the study/protocol

1836

E.6. Source of Animals - Please check the appropriate box(es) to indicate the source of the animals.

No answer provided.

F. Proposed Research - Comprehensive Summary

**** you can use the drop-down list above to temporarily skip past error messages. ****

F.1. Please add an experimental timeline (i.e., day 0, day 7, day 14, etc.) indicating the proposed treatment/procedure and the total duration of the experiment. Please coordinate the descriptions of experiments, experimental design, and timelines throughout the protocol.

Example might include 'time 0=begin experimental feeding, weeks 1/3/5=blood draw, week 10=euthanize animals'. Make sure to include all experimental manipulations.

This section should be a clear timeline of ALL procedures from start of experiment to euthanasia. See examples of flowcharts below or this example:

Day 1-Day 7: Daily drug X injections.

Day 8: blood draw/fecal collection

Day 9 to 69: Behavior testing on alternate days; fecal collection weekly

Day 70: Euthanize

F.1.1. It is encouraged that you attach a descriptive flowchart for clarity, especially for populations that undergo multiple procedures.

No answer provided.

Flowchart example 1

Flowchart example 2

F.2. For the summary of the CURRENT WORK, briefly explain the experimental design, then provide specific technical details about the proposed experiments for IACUC review.

- 1. Give a BRIEF experimental design statement for each experiment separately, including the techniques directly related to the use of animals such as breeding, research procedures, anesthesia, surgery, behavioral studies, pain relief (for both surgical and non-surgical procedures), euthanasia and experimental endpoints.**
- 2. For breeding protocols, indicate breeding scheme (monogamous or trio), weaning procedures, genotyping, etc.**
- 3. Specific details for ex vivo studies of tissue of cells removed from animals during surgery or following euthanasia are not needed.**
- 4. Do not discuss or justify the number animals to be used in this section. However, it is recommended that you identify the strains you will use in order to facilitate the review of your protocol, although this is not required. The idea is to provide scientific justification for use of multiple strains and to help the reviewers understand the experimental design.**
- 5. Please coordinate the descriptions of experiments, experimental design, and timelines throughout the protocol.**
- 6. Describe only the general goals of surgical procedures and their timing relative to other procedures.**

DO NOT CUT AND PASTE GRANT PROPOSAL LANGUAGE INTO THIS SPACE.

The protocol you submit is a "stand alone" document. Do not refer to procedures in other protocols or publications or assume that they are so generally understood or used that everyone will know what you will do. This section should correlate with what you included in the Experimental Timeline and should include a brief experimental design statements for each experiment. The committee does not require descriptions of in vitro experiments.

This section is for the reviewers. It is important that they understand the experimental plan in detail, so that they know what happens to each animal throughout the experiment, from beginning to end, and also that they understand how the experimental design accomplishes the goals of the research. Although it is not necessary to list all strains used in an experiment, sufficient detail should be given so that the reviewers (or readers) understand why a particular strain or strains are being used.

For breeding protocols, make sure you explain the breeding system (monogamous, trio breeding, etc.) as well as when and how you will wean the animals, genotype the animals, and tag or identify the animals.

For experimental protocols an example follows:

In this study, we propose to conduct an experiment to determine whether social stress will produce an escalation on ethanol consumption. We hypothesize that greater effects will be seen in FVB/NJ mice as we have seen for the intermittent access procedure.

Experiment 1: Social Defeat induced escalation of ethanol consumption in three different mouse strains (C57Bl/6J, DBA/2J, and FVB/NJ)

Methods and Design:

[list all methods here including amounts of compounds given/injected/etc., amounts of blood/urine/etc. taken and describe the overall experimental design]

F.3. Miscellaneous Attachments

No answer provided.

G. Animal Use

** you can use the drop-down list above to temporarily skip past error messages. **

G.1. This species was selected for the study because of the following attributes (select all that apply):

No answer provided.

G.2. The following is true of the sexes of animals used in the proposed study(s):

Entered: 10/14/20 **By:** Joseph, Elaine **Internal:** No

If only one sex is to be used (i.e., only females) you must justify that choice. If both sexes are to be used - this must be taken into account in Section E (Statistics) and accounted for this in your equations.

No answer provided.

G.3. Does this work unnecessarily duplicate any previous animal use either by your lab or published in the literature?

No answer provided.

G.4. Does this work contain any potentially painful and distressful procedures where relief (analgesia) is withheld, where pain is possible without analgesia, or death as an endpoint (unsedated death) is possible?

Entered: 10/14/20 **By:** Joseph, Elaine **Internal:** No

This should be checked "yes" only if you are performing painful procedures (such as surgery or wound creation) without analgesics or using death as an endpoint

No answer provided.

G.5. Will animals be transported between buildings on campus (i.e., on health science campus)?

Entered: 10/14/20 **By:** Joseph, Elaine **Internal:** No

This should be "yes" if you are moving animals between DLAR and another room, for example.

No answer provided.

Please see the [Animal Transportation Guideline](#) for more information.

G.6. Will animals be transported between campuses (Main Campus and Medical Campus) or off campus in a private (non-DLAR) vehicle?

No answer provided.

Please see the [Animal Transportation Guideline](#) for more information.

G.7. Please indicate all experimental or scientific deviations from Animal Welfare Standards or the Guide for the Care and Use of Laboratory Animals (check all apply):

No answer provided.

Exemption from temperature schedule: This includes exposure to extreme heat or extreme cold.

Exemption from standard lighting schedule: This includes exposure to 24 hour light or 24 hour dark.

H. Additional Information on Experimental Procedures

**** you can use the drop-down list above to temporarily skip past error messages. ****

H.1. Excluding surgery procedures or euthanasia, does this protocol have additional procedures such as genotyping, blood collections, injections, chemical or biological agent administration?

Entered: 10/14/20 **By:** Joseph, Elaine **Internal:** No

This should be checked "yes" if you are administering any biological or chemical agents to animals OR if you are taking blood, collecting urine/feces/tissue, or genotyping animals.

Yes

H.2. Do these procedures include the administration of drugs or chemical/biological agents into live animals?

Yes

If anesthetic is used for chemical restraint, list it among the agents.

H.2.1. Provide the following information for each agent that will be administered as part of this procedure. Each agent should be listed in a separate row. For each agent listed, provide the dosage (mg/kg body weight), the volume, the route of administration (IP, IM, IV, SQ, oral, topical), frequency and duration of administration (twice weekly for 5 weeks, etc.), and any known side effects of the administered agent. Include all paralytics and neuromuscular blocking agents. The rationale for the use of these agents must be described in previous section (Animal Use).

Agent	Dose/ Concentration	Volume	Route	Frequency/ Duration	Side Effects
ketamine xylazine	100 mg/kg 10 mg/kg	0.1ml	Intra- Peritoneal	once every 2 weeks for imaging, 30 minutes each session	intended to produce anesthesia

If you are unable to type in (locate) the agent in the drop down list, please contact the IACUC Office at IACUC@utoledo.edu with the exact spelling of your agent and whether it is a "Chemical" or "Biological" agent to have it added immediately. Please contact Elaine Joseph (419-383-4251) for any questions.

H.3. Are all of these Pharmaceutical grade agents?

Entered: 10/14/20 **By:** Joseph, Elaine **Internal:** No

This should be "no" if ANY of these agents are purchased from a chemical company (i.e., Sigma) and not from a pharmaceutical company such as Henry Schein or McKesson.

No answer provided.

Pharmaceutical grade agents are purchased from pharmaceutical companies such as Henry Schein. Non-pharmaceutical grade compounds include reagent or chemical grade and are purchased from companies such as Sigma.

H.4. Do these procedures require blood, body fluids, or tissue to be collected from live animals?

Yes

H.4.1. Provide the following information

Tissue/Fluid Type	Amount/Volume	Frequency	Collection Method
Blood for CBC	100ul	once a week	tail nick
Blood for GTT	10ul	0, 1, 3, 6, 12 hours	tail nick

H.5. List locations where experimental procedures will be performed. If multiple locations, explain what procedure will be performed at each location.

Entered: 10/19/20 **By:** Joseph, Elaine **Internal:** No

This should be DLAR, unless you will take your animals to the laboratory for blood draws, injections, etc. If you will perform experimental procedures in the laboratory (outside of DLAR), this requires IACUC approval.

No answer provided.

I. Surgery

** you can use the drop-down list above to temporarily skip past error messages. **

I.1. Does this protocol require surgery on live animals?

A Surgery involves an incision to open the skin/body cavity under anesthesia, whereas a Procedure may also involve anesthesia but no incision or opening of the skin/body cavity.

Yes

I.1.1. Provide the following information for each agent that will be administered for anesthesia, analgesia or sedation prior to any and all surgeries, during or after. Each agent should be listed in the table. Include all paralytics and neuromuscular blocking agents. For each agent, provide the drug name (prefer generic), dosage (mg/kg body weight), the route of administration (IP, IM, IV, SQ, oral, topical, etc.) and frequency/duration.

Entered: 10/14/20 **By:** Joseph, Elaine **Internal:** No

In this section, please list ALL anesthesia, sedatives, and analgesia for ALL survival and non-survival surgeries you will be performing.

Please see the IACUC website for Guidelines on Rodent Anesthesia and Rodent Analgesia

<https://www.utoledo.edu/research/rsp/RC/animal.html>

Drug	Dose/ Concentration	Route	Frequency/ Duration
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I.1.2. List all locations where any surgery will be performed (Campus, Building, Room#). If multiple locations, explain what surgeries will be performed at each location.

Entered: 10/14/20 **By:** Joseph, Elaine **Internal:** No

This should be DLAR. If you are performing surgeries outside of DLAR this requires IACUC approval.

No answer provided.

I.1.3. Will you be performing survival surgeries?

Yes

I.1.4. Will you be performing non-survival surgeries?

Yes

I.1.5. Will you be performing multiple survival surgeries on an individual animal (i.e., under separate anesthetic episodes)?

Entered: 10/19/20 **By:** Joseph, Elaine **Internal:** No

This should be 'yes' if one animal will undergo more than one survival surgery.

No answer provided.

J. Survival Surgeries (1 of 1)

**** you can use the drop-down list above to temporarily skip past error messages. ****

Describe EACH survival surgical procedure mentioned in section Animal Use.

Please refer to the DLAR Animal Surgery Guidance for accepted practice in surgeon and animal preparation.

J.1. Survival Surgery Name

No answer provided.

J.2. Surgery is performed with aseptic technique. Hair around surgical site is removed using surgical clippers. Gross debris is removed using alcohol and the surgical site is scrubbed three times alternating with chlorhexidine (or providone-iodine) followed by sterile water (or alcohol).

No answer provided.

Aseptic technique is employed to minimize infection and maintain sterility. It is required for all survival surgery. This involves proper preparation of the animal, proper sterile surgeon attire, sterilization of instruments, etc.

J.3. Ophthalmic ointment is applied to eyes.

No answer provided.

J.4. Sterile instruments will be used, along with sterile surgical drapes. Proper PPE will be used including sterile surgical gloves, surgical caps, surgical masks, and sterile gowns. If multiple surgeries are to be performed, instruments will be sterilized between surgeries using a glass bead sterilizer. A new sterile surgical pack will be used every five (5) procedures.

No answer provided.

Instruments should be autoclaved prior to use.

J.5. Describe surgical procedure from incision and ending with site closure. Specify the wound closure and specify when sutures will be removed. Also, specify any intraoperative support (monitoring, thermal support, fluids, etc.). Specify dosages of anesthetics and analgesics given prior and during surgery.

This should be a detailed surgical description that begins with the incision and ends with wound closure. Please include all surgical procedures, including thermal support given, supplement fluids given, etc. Specify dosages of anesthesia and analgesia given prior and during surgery.

You do not have to describe animal prep (i.e., surgical site prep, instrument sterilization, etc.), as those questions were answered previously.

Example mini-osmotic pump implantation:

Animals will be given buprenorphine (0.05 mg/kg, SQ) and then anesthetized with isoflurane (5% initially, reduced to 2% for the duration of the procedure) and placed in dorsal recumbency. The skin and instruments are prepared as described above. After adequate anesthesia and buprenorphine administration, a ventral incision will be made in order to separate skin, muscle, and fascia and gain access to the peritoneal cavity. Through the opening, a sterile osmotic minipump will be placed in the peritoneal cavity. The body wall incision will be closed by suturing (absorbable PDS sutures), then the skin incision closed using wound clips. For immediate relief of postoperative pain, bupivacaine (0.5% x 0.5mL), a local anesthetic will be applied at the site of incision. To prevent dehydration, 2 mL of saline/100g rat will be given subcutaneously.

J.6. Describe how animals will be monitored postoperatively and what methods will be employed to prevent infections and to alleviate post-surgical discomfort. If agents are to be administered, provide the dosage (mg/kg body weight) and route of administration (IP, IM, IV, SQ, oral, topical). Describe methods used to detect infections. If the procedure would be expected to cause pain/distress in a human, use of analgesia should be considered. Also, list the behaviors/signs that would indicate pain or distress in the animals. State the interval of post operative observations. Consult DLAR for guidance.

Example:

Animals will be monitored for signs of post-surgical pain and infection or other complications. Analgesics and good nursing care will be provided to reduce pain and distress. A SQ injection of buprenorphine (0.05 mg/kg) is given at the initiation of surgery to sustain the pain relief. Saline (2 mL of saline/100g) will be given subcutaneously to alleviate dehydration. Boost and food pellets will be placed on the floor of the cage for easy access. In addition, rats will be monitored a minimum of once a day for the first 7 days following the surgery, and 2x weekly for the duration of the study. To determine whether the animals have pain, distress, and infection, the PI or study personnel will specifically pay attention to the following parameters: For infection, drinking, eating, and walking patterns will be monitored. For pain, the presence of awkward gait, hunched back, ocular and/or nasal discharge, and aggressive behavior will be monitored. If any animal manifests signs of pain or infection, the veterinarian will be consulted and appropriate treatment provided.

**Click 'Add New Survival Surgery' to add an additional request.
Click 'Delete Page' to delete the page (this will delete the current page and all data on the current page)**

K. Non-Survival Surgeries (1 of 1)

*** you can use the drop-down list above to temporarily skip past error messages. ***

Describe EACH non-survival surgical procedure mentioned in section Animal Use.

Please refer to the DLAR Animal Surgery Guidance for accepted practice in surgeon and animal preparation.

K.1. Non-Survival Surgery Name

No answer provided.

K.2. Surgery is performed using clean technique. Hair around surgical site removed using surgical clippers. Gross debris is removed using alcohol. Instruments should be clean and gloves should be worn.

No answer provided.

K.3. Ophthalmic ointment is applied to eyes.

No answer provided.

K.4. Describe each surgical procedure from incision and ending with euthanasia of the animal.

No answer provided.

**Click 'Add New Non-Survival Surgery' to add an additional request.
Click 'Delete Page' to delete the page (this will delete the current page and all data on the current page).**

L. Euthanasia

** you can use the drop-down list above to temporarily skip past error messages. **

L.1. Describe specific behavioral and/or physiological criteria under which the experiment will be terminated early and animals will be euthanized due to excessive distress such as unmanageable seizures.

Example:

Any mouse displaying any of the following signs will be euthanized for humane reasons: poor body condition (body condition score <2); weight loss > 20%; labored breathing; signs of severe illness including scruffy, hunched and inactive; Self-mutilation; or any animal that in the opinion of the University Attending Veterinarian requires euthanasia.

L.2. How will animals be euthanized (check all that apply):

Entered: 10/19/20 **By:** Joseph, Elaine **Internal:** No

Please make sure that any euthanasia methods mentioned in F2 (Comprehensive Summary) are checked here. If you are going to be euthanizing pups (during breeding) younger than 10 days, this must be done through decapitation or cervical dislocation.

No answer provided.

L.3. How will death be assured via a secondary method (check all that apply):

No answer provided.

L.4. Location of Euthanasia (Campus, Building, Room#)

Entered: 10/14/20 **By:** Joseph, Elaine **Internal:** No

This should ideally be DLAR, unless you are euthanizing animals in your laboratory. If euthanizing in your laboratory, this requires IACUC approval.

No answer provided.

L.5. Will any animals on this protocol not be euthanized?

No answer provided.

Stage awaiting acceptance

