National Eye Institute **ANIMAL STUDY PROPOSAL AMENDMENT FORM** 6/2018	**ASP NUMBER** _____ **AMENDMENT APPROVAL DATE** _____

A. ADMINISTRATIVE DATA:

Principal Investigator: _____

Animal Study Proposal Title: _____

Amendment Title: Modifications to:

1. **Section F**, Description of Experimental Design and Animal Procedures, REPLACE the description of Rodent Tail biopsy procedure;
2. **Section I**, Anesthesia, Analgesia, Tranquilization, ADD potential treatment regime for Rodent Tail biopsy procedure and;
3. **Section J**, Method of Euthanasia, CHANGE the CO2 flow rate into the euthanasia chamber

B. ANIMAL REQUIREMENTS: **Check if No Additional Animals are Required** ☒

Species _____ Age/Weight/Size _____ Sex _____

Stock or Strain _____

Source(s) _____

Number _____

C. TRANSPORTATION AND HOUSING: **Check if No Change** ☒

(*This section must be addressed if there are any changes in transportation or housing.*)

D. PURPOSE OF AMENDMENT:

(*Briefly explain the purpose of the amendment and how the proposed changes fit the objectives of the original ASP.*)

E. JUSTIFICATION FOR NUMBER OF ANIMALS REQUESTED: **Check if No Change** ☒

(*You must provide numbers justification for any additional animals.*) 1) Explain your rationale for animal use.

2) Justify the appropriateness of the species selected. 3) Justify the number of animals to be used. 4) If applicable, justify why this study only uses animals of the same sex in all experimental groups. (Use additional sheets if necessary).

F. 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.)

Replace the description of Rodent Tail biopsy procedure with:

The NEI Rodent Tail biopsy policy will be followed. Alternatively, tail biopsies may be conducted by Technical Request, by the Animal Facility Holding Personnel **in accordance with their Facility's ACUC-approved SOPs.**

G. SURVIVAL SURGERY:

Check if No Change ☒

(Describe all additional or changes in surgical procedures to be performed. Include information on aseptic technique, identification of individuals conducting procedures, post-operative care and monitoring, and location. If multiple survival surgeries are proposed, the total number of surgeries must be indicated clearly; this includes cases where animals will have undergone surgery on previous studies.)

H. PAIN OR DISTRESS CATEGORY:

Check if No Change ☒

(List the category for new animals to be added under amendment. IF ANIMALS ARE INDICATED IN COLUMN E, A SCIENTIFIC JUSTIFICATION IS REQUIRED. PLEASE COMPLETE THE EXPLANATION FOR COLUMN E LISTINGS FORM AT THE END OF THIS DOCUMENT. THIS FORM WILL ACCOMPANY THE NIH ANNUAL REPORT TO USDA.)

Category	Number of Animals
☐ ***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	

(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 paragraph 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 databases (2 or more) searched, the date of the search, period covered, and keywords used. Keywords must relate to the procedures and treatments with a potential to cause pain and/or distress. Please indicate the procedure or treatment which resulted in an animal being placed in Columns D or E.)***

I. ANESTHESIA, ANALGESIA, TRANQUILIZATION: Check if No Change ☐

(For animals indicated in Section H, Column D, specify the anesthetics, analgesics, sedatives or tranquilizers that are to be used. Include the name of the agent(s), the dosage, route and schedule of administration. All substances administered to an animal, except those routinely considered as food, must be pharmaceutical grade unless otherwise justified in accordance with the NIH ARAC Guidelines for the Use of Non-Pharmaceutical Grade Compounds in Laboratory Animals. and as such may need to consider factors such as grade, purity, sterility, acid-base balance, pyrogenicity, osmolality, stability, site and route of administration, compatibility of components, side effects and adverse reactions, storage, and pharmacokinetics.)

For tail biopsies of approximately 0.2 cm of the terminal tail in mice or rats between 10-28 days of age:

- Topical anesthesia: regional hypothermia using ice-cold ethanol
- Lidocaine-Epinephrine-Tetracaine gel (L.E.T.) (4%/0.05%/0.5%, Wedgewood Pharmacy),

For tail biopsies of approximately 0.2 cm of the terminal tail in mice or rats >28 days of age:

- General anesthesia:
  - Isoflurane 1-3%, to effect;
  - Ketamine/xylazine, 70-100 mg/kg Ket/5-10 mg/kg Xyl
- L.E.T. gel
- a single dose of a systemic analgesic

  For Mice:

  - Meloxicam: Mice mg/kg BW SQ or PO q24hr
  - Carprofen, 5 mg/kg BW, SQ or PO q24hr
  - Ketoprofen, 5 mg/kg BW, SQ q24hr
  - Buprenorphine, 0.05 mg/kg BW, SQ q24hr

  For Rats:

  - Meloxicam: 2 mg/kg BW SQ or PO q24hr;
  - Carprofen, 5 mg/kg BW, SQ or PO q24hr
  - Ketoprofen, 5 mg/kg BW, SQ q24hr
  - Buprenorphine, 0.05 mg/kg BW, SQ q24hr

- If oral dosing via the water bottle is preferred, Carprofen 0.025 mg/ml drinking water may be used for both rats and mice.
- Alternatively, Buprenorphine sustained release (SR) 0.6 mg/kg BW SQ for mice and 1.3 mg/kg BW SQ for rats, will provide up to 72 hours of analgesia.

For mice or rats of any age requiring a second tail biopsy or a biopsy of >5 mm

- General anesthesia, either with isoflurane or ketamine/xylazine
- L.E.T. gel
- systemic analgesic for one to three days post biopsy, see list above

J. METHOD OF EUTHANASIA OR DISPOSITION OF ANIMALS AT END OF STUDY:

(Indicate the proposed method, and if a chemical agent is used, specify the dosage and route of administration. If the method(s) of euthanasia include those not recommended by the AVMA Guidelines on Euthanasia, provide justification why such methods must be used. Indicate the method of carcass disposal if not as MPW.)

Check if No Change ☐

Based on the new AVMA Guidelines for the Euthanasia of Animals: 2020 Edition effective 1-15-2020, change the CO2 flow rate into the euthanasia chamber **from** 10-30% of cage volume/min **to** 30-70% of cage volume/min.

K. USE OF HAZARDOUS AGENTS:

Check if No Change ☒

(Use of hazardous agents requires the approval of an IC safety specialist.)

Biological Agents with Pathogenic Potential: **NONE** ___ (check if none) For guidance, see ORS/DOHS Biological Safety and Compliance**.** Include the NIH Institutional Biosafety Committee's risk-assessment language or attach a copy of the registration documents.		
Agent:	PRD #:	ABSL:
Additional occupational health and/or animal facility handling safety considerations.		

Recombinant DNA: **NONE** ___ (check if none) For guidance, see NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules FAQs. Include the NIH Institutional Biosafety Committee's risk-assessment language or attach a copy of the registration documents.		
Recombinant DNA:	RD #:	ABSL:
Additional occupational health and/or animal facility handling safety considerations.		

Ionizing Radiation: (Radionuclides & radiation producing equipment) **NONE** ___ (check if none) For guidance, see ORS/DRS/Policies/Radiation Safety Protocols Animal Studies Proposal Requirements
☐ Yes, I will use radionuclides or radiation producing equipment as part of the experimental procedures on the ASP and all operators will be registered with Division of Radiation Safety. If an irradiator is to be used, then all individual users must comply with Division of Radiation Safety requirements for irradiator training, and all individual assessors will comply with applicable security requirements for escorts and proxy card access approval.
List of Radionuclides:
Radiological safety considerations:

Hazardous Chemicals or Drugs: **NONE** ___ (check if none) For guidance, see NIH Policy Manual 3034 – Working with Hazardous Chemicals Material safety data sheets for hazardous chemicals and drugs must be maintained readily accessible to laboratory and animal facility employees (Title 29, Part 1910.1200(b)(3)(ii), CFR)
List of Agents:
Additional occupational health and/or animal facility handling safety considerations:

L. BIOLOGICAL MATERIAL/ANIMAL PRODUCTS FOR USE IN ANIMALS:

(List cells/tissues, sera/antibodies, viruses/parasites/bacteria, and non-synthetic biochemicals that will be introduced into research animals.)

Check if No Change ☒

Material:	Source:	Sterile?	
		Y	N
If derived from rodents, has the material been tested, e.g. MAP/RAP/HAP/PCR? (If Yes, attach copy of results)			
Have the tested materials been passed through rodents outside of the animal facility in question?			
Is the material derived from the original MAP/RAP/HAP/PCR tested sample?			
I certify that to the best of my knowledge that the above is complete and correct, and that the material remains uncontaminated with rodent pathogens.			

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.), safety precautions, or monitoring. Include justification for exemption from participation in the environmental enrichment plan for nonhuman primates or exercise for dogs.) None ☒

N. CONCURRENCES:

Principal Investigator:

Name _____Signature _____Date _____

Laboratory/Branch Chief, certification of review and approval on the basis of scientific merit and sex as a biological variable. (Note: Scientific Director's signature required for proposals submitted by a Laboratory or Branch Chief):

Name _____Signature _____Date _____

Safety Representative, certification of review and concurrence (Required of all studies utilizing hazardous agents):

Name _____Signature _____Date _____

Radiation Safety Representative, certification of review and concurrence (Required of all studies utilizing radioactive materials):

Name _____Signature _____Date _____

Facility Manager, certification of resource capability in the indicated facility to support the proposed study:

Name _____Signature _____Date _____
COMMENTS:

Facility Veterinarian, certification of review:

Name _____Signature _____Date _____

Attending Veterinarian, certification of review:

Name _____Signature _____Date _____

N. FINAL APPROVAL:

NEI Animal Care and Use Committee Chairperson, certification of review and approval:

Name _____Signature _____Date _____