UNIVERSITY OF WISCONSIN-MADISON SCHOOL OF MEDICINE AND PUBLIC HEALTH BIOMEDICAL RESEARCH MODEL SERVICES

STANDARD OPERATING PROCEDURE

NUMBER: 610 EFFECTIVE DATE: 12/1/18

TITLE: Genotyping and Identification Procedures for Mice

SUPPORTING DOCUMENTS:

PURPOSE:	To establish a procedure for genotyping, and identification procedures for mice.	
SCOPE:	All mice.	
RESPONSIBILITY:	It is the responsibility of breeding core staff to report any injuries, illnesses or deaths of mice. Breeding core staff must take care of animal safety.	
SAFETY:	Follow Personal Protective Equipment (PPE) Requirements (SOP 600) when entering any animal/procedure rooms. Read directions on label and/or SDS for all cleaning and sanitizing agents.	
TRAINING REQUIREMENTS:	Careful reading of related SOPs and hands on training	

MATERIALS NEEDED: 2 mm ear punch, iris or suture scissors, bead sterilizer, forceps, 70% Alcohol, Eppendorf tube or well plate, Fisher Chemical resistant pen, bags, disinfetant and Innovive cage.

PROCEDURE:

1. Ear Punching

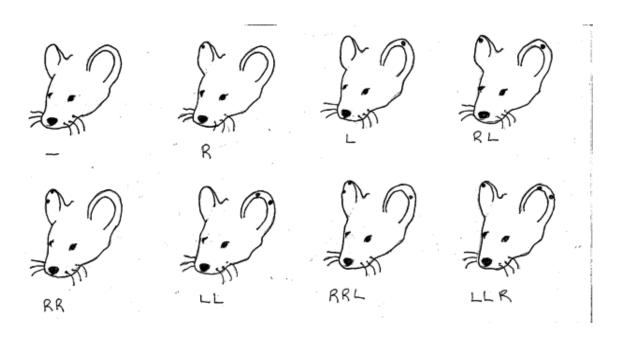
- **1.1.** Mice must be <u>14 days or older</u> of age and have ears large enough to place two ear punches. Ears should be thin at this point.
 - **1.1.1.** If uncertain that they are large enough recheck in a few days.
- **1.2.** Record cage number in cage side assistant.
 - **1.2.1.** Click enter progeny
 - **1.2.2.** Record number of pups and sexes
 - 1.2.3. Record ear punch under code and add coat color
 - **1.2.4.** Check sample mice for genotyping. (NOTE** If you are not weaning them make sure the wean box is unchecked)
- **1.3.** Ear tissue can also be used for PCR, when requested by the lab.
- **1.4.** Label Eppendorf tube with animal ID. Use the chemical pen so that it can't wipe off during processing.
- **1.5.** When collecting samples for PCR, dip the ear punch in 70% alcohol between samples
- **1.6.** Spray the outside of cage with disinfectant before placing in the hood.
- 1.7. Set up an empty cage to hold mice once they have been ear punched.

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1.8. When selecting animals to be ear punched it helps to choose animals of different color, when available, for similar punches (ex. R/black vs. R/agouti). This will help ID animals in the case you cannot read an ear punch.

- 1.9. Keeping your gloves wet with disinfectant, restrain each mouse and put a punch in the ear. See punch system below:
 - 1.9.1. Separate by sex.
 - **1.9.2.** Both ears no punches
 - **1.9.3.** Right ear 1 punch
 - **1.9.4.** Left ear 1 punch
 - **1.9.5.** Right ear 1 punch and left ear 1 punch
 - **1.9.6.** Right ear 2 punches
 - **1.9.7.** Left ear 2 punches
 - **1.9.8.** Right ear 2 punches and left ear 1 punch
 - **1.9.9.** Right ear 1 punch and left ear 2 punches
 - **1.9.10.** Right ear 2 punches and Left ear 2 punches
- 1.10. If collecting samples for PCR, place tissue sample in tube. You can use a forceps when collecting ear punches. Close cap
- 1.11. Place samples in small plastic bag. Label strain and PI name on outside of bag. Keep strains separate. If collecting multiple strains, place small bags in larger bag labeled with PI name.
- **1.12.** Record an "X" on the sticker dot on the cage to signify samples have been collected. Make sure all punches have been recorded under "code" in Mosaics.



2. Tail snips

- **2.1.** Mice should be sampled at 14-21 days.
- 2.2. Occasionally some strains of mice are slower growing but have to be less than 21 days to collect tails without anesthesia.
- **2.3.** Spray the outside of cage with Virkon S before placing in the hood.

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2.4. Set up on empty cage to place mice once they have been ear punched and tail snipped.

- **2.5.** Keeping your gloves wet with disinfectant, restrain each mouse and put a punch in the ear. (See punch system above.)
- **2.6.** Cut less then 5mm of the tip of the tail off with a hot scissors that has been in the bead sterilizer.
 - **2.6.1.** Scissors should be hot to cauterize tail.
 - **2.6.2.** Replace scissors in bead sterilizer to allow for complete sterilization between each tail snip so that scissors does not cause DNA contamination between mice.
- **2.7.** Place tail snip in sealed tube, label tube with mouse id.
- **2.8.** Place samples in small plastic bag. Label strain and PI name on outside of bag. Keep strains separate. If collection multiple strain, place small bags in larger bag labeled with PI name.

NOTE: Record an "X" on the sticker dot on the cage to signify samples have been collected. Make sure all punches have been recorded under "code" in Mosaics

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Revision	Revision Description	Effective Date
0	Initial Release	August 1, 2017
1	Revision following transfer to SMPH	December 1, 2018