UNIVERSITY OF DELAWARE
OFFICE OF LABORATORY ANIMAL MEDICINE

Tissue Collection for Genotyping SOP #PRO-009

Samples may be collected from genetically-modified rodents to determine genotype. Tissue biopsies may be collected from the tail, pina, or distal phalanx. Recently, non-invasive methods using hair follicles, blood, feces, tears, or oral swabs have been described. The least invasive method that is practical should be used.

Sample collection should be done in a manner that minimizes the risk for cross contamination.

Tail Biopsy

Performing tail biopsy on mice that are as young as possible to minimize potential pain.

Anesthesia and analgesia:

• not required for animals prior to weaning (21 days).

• required for animals older than 21 days or for any animal that undergoes a repeat tail biopsy

• Anesthetics and analgesics should be chosen in consultation with the Attending Veterinarian and follow SOP F-008 Protocol for using isoflurane anesthesia machine, and SOP A-103 Use of buprenorphine in mice if applicable .

• Local anesthesia may be provided by immersion of the tail tip in ice cold ethanol for 10 seconds for rodents less than 28 days.

• General anesthesia with isoflurane is acceptable for all ages.

• Post-procedural analgesia (such as topical analgesics or buprenorphine) should be considered and the need will increase with the age of the rodent or with repeat biopsies.

Sample size:

• Tail biopsy should be the smallest amount possible. Typically, 2mm is sufficient.

• Amounts greater than 2mm must be justified in the protocol and approved by the IACUC

Hemostasis:

The animal must be monitored to assure hemostasis after tail biopsy. If needed, acceptable methods of hemostasis include digital pressure, brief heat cautery to an animal under general anesthesia, or styptic powder.

Equipment Required:

• Gloves

• Sterile sharp scissors or sterile scalpel blade (If using a bead sterilizer, allow it to heat to manufacturer recommended temperature)

• 70% alcohol to clean work surface and tail (providone-iodine or chlorhexidine solutions may interfere with DNA analysis)

• Cauterization method (manual pressure with clean gauze or hemostatic agent such as styptic powder)

Technique:

1. Carefully restrain the animal. (Anesthetize if required or as necessary.)

2. Cut the tip of the tail using sterile scissors or sterile blade.

3. Scissors should be sterilized with 70% alcohol or a bead sterilizer between animals. The scissors then must be allowed to cool prior to the tail snip.

4. Control bleeding using digital pressure or cauterization.

5. Make sure tail is not bleeding before returning animal to a clean cage. If it is still bleeding, contact the veterinary technician or the veterinarian.

6. Verify the animal's ID and confirm that it matched the sample tube.

7. Observe animal for bleeding or any signs of pain or distress such as biting at the tail. If signs are observed notify the Attending Veterinarian.

8. Check animal daily to ensure tail is healing. If the tail is not healing as expected or appears infected, notify the Attending Veterinarian.

Pinna biopsy

Pinna biopsy may allow tissue collection and identification of the animal. It is suitable for rodents 14 days or older. A two millimeter ear punch or notch is recommended, and the punch should be sterilized with a bead sterilizer or cleaned with 70% ethanol between animals. The use of analgesics or anesthetics is not required.

Technique:

1. Carefully restrain the animal.

2. Cut the ear using sterile scissors or use a sterile ear punch.

3. Scissors or the ear punch should be sterilized with 70% alcohol or a bead sterilizer between animals. The equipment must be allowed to cool prior to use.

4. Check animal daily for the next week. If the ear is not healing or appears infected, notify the Attending Veterinarian.

5. Verify the animal's ID and confirm that it matched the sample tube.

Distal Phalanx Biopsy

Removal of the portion of the digit, distal phalanx biopsy, may be used to identify rodents and to obtain tissue for genotyping. This technique should only be used in mice and rats after the digits are no longer webbed and before they reach 8 days of age. Only one digit should be used and it is preferrable to use the hind paw. If the forepaw must be used, it is preferrable not to use the hallux (dew claw). Anesthetic or analgesics are not required.

Technique:

1. Carefully restrain the animal.

2. Cut the toe using sterile scissors.

3. Scissors should be sterilized with 70% alcohol or a bead sterilizer between animals. The equipment must be allowed to cool prior to use (minimum time for cooling at least one minute).

4. Check animal daily for a week. If the toe is not healing as expected or appears infected, notify the Attending Veterinarian.