

PROTOCOL- Harvest Tail Biopsies for gDNA Isolation

Tail biopsies from putative transgenic animals or from animals in an existing transgenic colony can be taken from animals up to 2 weeks of age without administering analgesic or anesthesia. After 2 weeks of age analgesic and anesthesia must be administered prior to initiating a biopsy. In general, animals are given an identifying mark at the time of biopsy. The Transgenic/Knockout Core Facility uses toe tattoos to identify individuals at the time of biopsy. See the attached chart for identification of toe tattoo marks.

Materials: List of animals and labeled eppendorf tubes (1.5 mL)
Tattoo needle holder (with magnification)
30G1/2 hypodermic needle
1 mL syringe plunger (to hold paste)
Small dissecting scissor
Eppendorf tube containing soapy water (to rinse scissor between animals)
Kimwipe or paper towel
Ketchum Tattoo Ink Paste – green (www.ketchum.ca)

Method:

1. Set up the tattoo station: Spray the hood with disinfectant and apply a wet spot in one side of the hood. Place a paper towel over the wet spot. Place the labeled tubes in a tube rack on the paper towel and insert the syringe plunger into one hole of the rack. Apply a spot of tattoo ink to the upright tip of the plunger. Open the scissors and rinse with soapy water, wipe off with a Kimwipe and set to one side (on the wetted paper towel).
2. Remove pups from cage and check for sex, color, etc. It will be easier later to read toe tattoos if pups are separated into groups according to sex, color, etc. prior to tattooing, if possible. If marks are not needed proceed to the biopsy, step 5.
3. Remove a pup and hold it in your left hand (upside down) holding the right hind leg between your left thumb and forefinger. It should be possible to hold the leg close to the body so the toes are well extended.
4. With the toes of the right hind leg extended hold the tattoo needle (in holder) in your right hand. Touch the needle tip to the spot of ink on the plunger and position it over the extended toes. Carefully touch the needle to the appropriate toe and squeeze the ink holder until the tip of the needle pierces the designated toe. Some people find it easier to have the needle tip go through the hole on the tattoo holder, others prefer it to touch the hard surface on the edge of the holder. Set it up the way that is easier for you. The needle will pierce the toe and travel through it to give a good, permanent mark. Work neatly and you will be able to see your marks.
5. Once marked keep the same hold on the pup and position it over the designated collection tube, using clean dissecting scissors remove the tip of tail (~2-3mm) and allow the tail tip to fall into the collection tube. Check the tube for the harvested tissue. When finished the mouse can be returned to the cage.
6. Carefully rinse the scissor between animals using soapy water and wipe clean.

7. Harvested tissue should either be processed immediately or frozen until needed.
8. Animals should be checked frequently for full recovery from the biopsy procedure. Animals under 2 weeks of age rarely bleed from biopsies however if bleeding occurs an appropriate styptic should be applied (i.e. 3M VetBond, 3M Animal Care Products #1469SB). A tissue adhesive should always be applied to older animals when biopsied.

