



Genotyping Procedures

Genotyping

The proper identification of genetically engineered animals is critical to your research and in reducing the number of animals involved in a research project. There are many types of biological samples that can be used for genotyping your transgenic animals:

- Fin Clips (Fish)
- Tail Biopsy
- Ear Punch
- Fecal Pellet
- Hair Sample
- Blood Sample

Rodents are generally genotyped and identified before or at weaning. The [NIH ARAC Guidelines for Genotyping Mice & Rats](#) provides specific information and references about how and when genotyping procedures should be performed.

Caudal Fin Clip

Zebrafish will regenerate caudal fin tissue that is removed. Caution must be used to only remove the fin tissue, and not damage the peduncle or bleeding and permanent tissue damage may occur.

- Anesthetize the fish in MS-222.
- With a sterile scalpel or scissors, cut up to 1/2 to 2/3 of the caudal fin.
- Use caution not to cut into the peduncle tissue.
- Recover the fish in fresh system water.



Tail Biopsy

The [NIH ARAC Guidelines for the Genotyping of Rodents](#) recommends that rodent tail biopsies be performed at 10 – 21 days of age before the vertebrae are calcified.

- **Mice & Rats 10 – 21 days of age** - - a local anesthetic such as ethyl chloride or ice cold ethanol should be used.
- **Mice & Rats >21 days of age** - a local anesthetic such as ethyl chloride or ice cold ethanol or a general anesthetic should be used.
- **Rats >35 days of age** – general anesthetic is required.

1. Restrain the animal by grasping the skin along its back with your left hand (if right-handed).
2. Clean tail with alcohol.
3. Apply topical anesthetic.
4. With a sterile scalpel, razor blade, or scissors, cleanly excise the distal 2 – 5 mm of the tail.
5. Apply gentle manual pressure or silver nitrate to stop bleeding.
6. At this time, the animal should be identified by an appropriate method (ear tag, transponder, ear punch,...)



Ear Punch

Placement of the ear punch hole can be used to identify animals as well as obtain a genotyping sample.

1. Restrain the animal by grasping the skin along its back with your left hand (if right-handed).
2. Place head of ear punch in desired location on the mouse ear pinna as shown in middle picture.
3. Firmly & quickly squeeze the punch to create a clean hole in the ear as shown in picture on right.



Fecal Sample

Fresh fecal samples can easily be obtained by removing animals from their home cage, and gently restraining the animal until it defecates. Most mice will defecate within a minute of being picked up. Caution should be used to avoid cross-contamination of samples. At this time, the animal should be identified by an appropriate method (ear tag, transponder, ear punch,...)

Hair Sample

Hair bulbs may be collected and used for polymerase chain reaction (PCR). Only a few hair bulbs per animal should be necessary to obtain adequate DNA.

- Restrain the animal by grasping the skin along its back with your left hand (if right-handed).
- Pluck a small tuft of hair from the body using a pair of sterile forceps. A fresh pair of forceps must be used for each animal to avoid cross-contamination of the sample.
- At this time, the animal should be identified by an appropriate method (ear tag, transponder, ear punch,...)

