



## TISSUE SAMPLING POLICY FOR RODENT GENOTYPING

### Overview/Purpose

Analysis of rodent DNA may be necessary to verify the genetic composition of individual animals. Common collection sites include tail-snip/biopsy and ear punch/notch although hair roots, fecal pellets, and buccal swabs are less invasive and should be considered if very small quantities of DNA could be used for genotyping.

### Requirements

Tissue sampling for genotyping should be done as early as possible in the lifespan of the mouse to minimize the potential for pain/distress. Expectations for the common collection sites are as follows:

- **Ear Punch/Notch Procedure:** Manual restraint without anesthesia is acceptable. The ear punch/notch can also serve as a method of individual animal identification. The amount of tissue collection using this procedure is limited, and care should be taken not to accidentally drop/lose the ear tissue.
- **Tail-Snip/Biopsy Procedure:** When tail tissue (<5mm) is collected in mice up to and including 21 days of age, anesthesia is not required. General anesthesia is however required if removing greater than 5 mm of tissue (regardless of age) or if performing a tail-snip/biopsy in rodents 22 days or older\*. In all cases, hemostasis must be accomplished using digital pressure or styptic powder (Kwik-Stop®). If the animal is anesthetized, cautery may be used for hemostasis via a cautery pen, or via heated instruments used to collect the sample (scissors or blade heated in a hot bead sterilizer).

Tissue sampling must be performed using sharp devices to avoid tissue crushing. Instruments must be cleaned between individual animals to minimize cross-contamination of genetic material. Animals must not be returned to the home cage until hemostasis is achieved.

\* Tail-snip/biopsy of mice 22 days or older must be described as a protocol activity and approved by the IACUC

### Additional Resources

1. Boivin et al, 2016. A highly efficient strategy to determine genotypes of genetically-engineered mice using genomic DNA purified from hair roots  
<http://journals.sagepub.com/doi/abs/10.1177/0023677216646088>
2. Bonaparte D, Cinelli P, Douni E, Haurault Y, Maas M, Pakarinen P, Poutanen M, Lafuente MS, Scavizzi F. 2013. FELASA guidelines for the refinement of methods for genotyping genetically-modified rodents: a report of the Federation of European Laboratory Animal Science Associations Working Group. Lab Anim. Jul;47(3): 134-45.
3. Boivin, GP, et al. 2013. Genotyping DNA Isolated Using Cross-Linked Iminodiacetate Styrene Divinylbenzene Copolymer Beads. J Am Assoc Lab Anim Sci 52:682.
4. Braden-Weiss, G.C., Brice, A.K., Hankenson, F.C. 2011. Minimizing the impact of tail

biopsy in preweanling laboratory mice: inhaled isoflurane compared with topical anesthetics. J Am Assoc Lab Anim Sci 50: 736-737.

5. Broome, R. L., L. Feng, Q. Zhou, A. Smith, N. Hahn, S. M. Matsui, and M. B. Omary. 1999. Non-invasive transgenic mouse genotyping using stool analysis. FEBS Letters 462:159-60.
6. Burkhart, C. A., M. D. Norris, and M. Haber. 2002. A simple method for the isolation of genomic DNA from mouse tail free of real-time PCR inhibitors. Journal of Biochemical and Biophysical Methods 52:145-9.
7. Cinelli, P., A. Rettich, B. Seifert, K. Burki, and M. Arras. 2007. Comparative analysis and physiological impact of different tissue biopsy methodologies used for the genotyping of laboratory mice. Laboratory Animals 41:174-84.
8. Garzel LM , Hankenson FC, Combs J, Hankenson KD. 2010. Use of quantitative polymerase chain reaction analysis to compare quantity and stability of isolated murine DNA. Lab Anim (NY), 39(9): 283-289.
9. Hankenson, F.C., Braden-Weiss, G., Blendy, J.A. 2011. Behavioral and activity assessment of laboratory mice (*Mus musculus*) after tail biopsy under isoflurane anesthesia. J Am Assoc Lab Anim Sci 50:686-94.
10. Hankenson, F. C., L. M. Garzel, D. D. Fischer, B. Nolan, and K. D. Hankenson. 2008. Evaluation of tail biopsy collection in laboratory mice (*Mus musculus*): vertebral ossification, DNA quantity, and acute behavioral responses. J Am Assoc Lab Anim Sci 47:10-8.
11. Jones CP, Carver S, Kendall LV. 2012 . Evaluation of common anesthetic and analgesic techniques for tail biopsy in mice. J Am Assoc Lab Anim Sci. Nov;51(6):808-14.
12. Paluch, LR, Lieggii, CC, Dumont, M, Monette, S, Riedel, E, Lipman, NS. 2014. Developmental and Behavioral Effects of Toe Clipping on Neonatal and Prewanling Mice with and without Vapocoolant Anesthesia. JAALAS Vol 53(2): 132-140.

## History of Revisions

**014-00** - Original policy; approved 02/25/2005

**014-01** – Revised to include requirements for tail snips at a given age; approved 06/22/2007

**014-02** – Revised to reflect requirements for exceptions to the policies; approved 04/22/2011

**014-03** - Policy revisions reflect the requirements for common DNA collection sites; approved 08/15/14

**014-04** – Policy was renamed, former title was Rodent Genotyping Policy. Also revisions reflect the requirements for accomplishing hemostasis, use of a cautery pen, and tail-snip/biopsy of mice 22 days or older must be approved as a protocol activity; approved 03/17/17