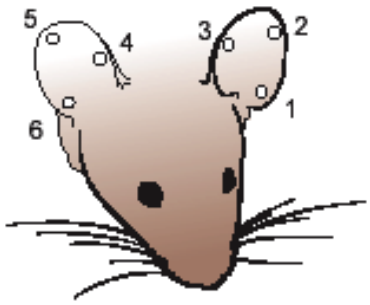
NHGRI ANIMAL CARE AND USE COMMITTEE (ACUC) GUIDELINE AND PROCEDURES FOR TAIL BIOPSY FOR DNA ANALYSIS AND GENOTYPING IN MICE

General Considerations

The NHGRI ACUC has determined that, even in the hands of a properly trained investigator, tail biopsy causes transient pain and distress. Investigators should be aware of this and consider alternatives.[1] The NHGRI ACUC has also determined that tail biopsy, performed with the modifications described below, is a safe, aseptic, and humane method of obtaining tissue from a mouse. DNA prepared from tail biopsies is suitable for analysis by either Southern Blot or PCR analysis. Procedures for isolating DNA from mouse tail biopsies can be obtained from the NHGRI Transgenic Mouse Core.

Guidelines

- Tail biopsy for DNA analysis and/or genotyping must be approved in the Animal Study Proposal.
- The ideal time to collect tail tissue is 10 to 15 days of age. At this age, the tail tissue is soft and the yield of DNA is highest. In addition, prompt analysis of tail tissue allows the desired mice to be identified prior to weaning which can save valuable space in the animal facilities. Tail snips can be performed on animals greater than 15 days of age but will produce a lower yield of DNA. The same procedures are utilized as for the animals greater than 10 days of age.
- Manually restrain mouse between thumb and forefinger. Identify mouse.[2] Refer to diagram below for NHGRI ear punch numbering scheme. Any combination of the numbers shown below can be used to generate numbers greater than 6.
- Mice greater than 10 days of age must be treated with a topical hypothermic (e.g., ethyl chloride)[3] or anesthetized[4] prior to collection of tail tissue.
- Wipe tail with 70% alcohol and allow to dry.
- With sterile scissors, cleanly excise the distal 5 mm of tail. If the proper procedures are followed, the yield of DNA from 5mm of tail should exceed 50 micrograms, enough for multiple analyses. Wipe scissors with 70% alcohol between animals.
- Assure hemostasis. Apply digital pressure, silver nitrate, or other cautery, if necessary, to control bleeding.
- There are currently no approved animal study proposals that incorporate repeat tail biopsies. Repeat tail biopsies, necessitated by laboratory error or other reasons, require the notification of the APD and supervision by the NHGRI Transgenic Mouse Core Facility personnel. The ARAC Guideline on genotyping http://oacu.od.nih.gov/ARAC/FinalGenotyping0602.pdf requires general anesthesia for repeat tail biopsies but the NHGRI ACUC accepts the use of ethyl chloride since it is recommended for use as a topical anesthesia for minor surgical procedures. NHGRI also requires that Lidocaine cream be applied following a repeat tail snip.



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[1] Chen, S. and G. Evans. A Simple Screening Method for Transgenic Mice Using the Polymerase Chain Reaction. Biotechniques. 8:32-33. 1990.
Amendola, R., S. Haendel, *et al*. Transgene Integration in Hair Follicles and Peripheral Blood Cells Measured by *In Vitro* DNA Amplification and Fluourescence *In Situ* Hybridization. DNA & Cell Biol. 10:311-317. 1991.
Schmitteckert, EM, C. Prokop, *et al*. DNA Detection in Hair of Transgenic Mice – a Simple Technique Minimizing the Distress on the Animals. Lab. Anim. (UK). 33:85-389. 1999.
Irwin, MH, R. Moffat, *et al*. Identification of Transgenic Mice by PCR Analysis of Saliva. Nat. Biotech. 14:1146-1148. 1996.
Hofstetter, JR, A. Zhang, *et al*. Genomic DNA from Mice: A Comparison of Recovery Methods and Tissue Sources. Biochem. & Mol. Med. 62:197-202. 1997.
[2] See Investigator Handbook and NHGRI Guideline 01.3 for Identification Methods
[3] Your veterinarian will supply you with ethyl chloride and advice on its proper use.
[4] See Isoflurane Standard Operating Procedure in Investigator Handbook