

THE UNIVERSITY OF MICHIGAN
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MEMORANDUM/SOP

TO: ULAM Veterinary Faculty, Postdoctoral Fellows, Veterinary Technicians,
Animal Care Staff, University of Michigan Research Faculty

FROM: F. Claire Hankenson, DVM, MS

DATE: November 21, 2005

SUBJECT: Tail Biopsy in Mice

PURPOSE: To provide guidelines for tail biopsy (cutting off the distal portion of the tail) taken for murine genotype analysis. The information represents a polled consensus among institutions that work with transgenic and mutant mice.

GUIDELINES:

1. In order to obtain 50-100 mg of high molecular weight DNA, 10 to 15 mm of tail from a 2-3 week old mouse is sufficient (1). The tail clip or biopsy procedure is momentary, but involves bone or cartilage, blood vessels, nervous tissue and skin. There is potential for pain from this procedure. Analgesics may be used if prolonged pain is anticipated. The smallest possible section of tail should be removed and adequate hemostasis should be achieved. The UK Joint Working Group on Refinement recommends no more than 5 mm of the tail for biopsy, and discourages repeated tail sampling (7).

2. In the mouse, the distal tail is completely ossified and innervated between 2 to 4 weeks of age. Thus, tail sampling is recommended in mice less than 3 weeks of age to avoid undue stress and discomfort to the animals. The optimum age at which to perform biopsy is between 12 to 16 days (University of Minnesota, Washington University, UCLA).

3. If the mouse is greater than 21 days of age, anesthesia is used for any amount of tail removal (Jackson Laboratory). Tail biopsy requires only brief anesthesia, and ULAM recommends inhalant agents, such as isoflurane, in an open-drop technique (300ul of liquid isoflurane, impregnated onto gauze, maintained in a 0.5 L jar). The animal cannot come into direct contact with the liquid anesthetic agent. Alternatively, mice can be anesthetized using isoflurane with a precision vaporizer and a nose cone. If fume hoods are unavailable for anesthetic scavenging, injectable anesthetics can be used. Topical anesthetics, e.g. lidocaine or ethyl chloride spray, are also acceptable but their efficacy at relieving pain associated with tail biopsy is unproven. Hence their use should be limited to those situations in which isoflurane

cannot be used. These situations need to be scientifically justified in the animal use application and approved by UCUCA.

4. Any instrumentation that is used to perform the biopsy should be sharp and sterile. This can be accomplished by using disposable scalpel blades or razor blades. Please note that disposable scalpel blades are not designed to be used on multiple animals, and a fresh blade must be used for each mouse. Alternatively, scissors can be sterilized using a hot bead sterilizer or disinfected using a cold-sterilizing liquid (Clidox, Sporocidin, etc) for a prolonged contact time (at least 15-60 minutes).

5. Hemostasis of the tail biopsy site can be achieved using compression, tissue adhesives (ex. Nexaban), styptic pencils, silver nitrate, or cautery.

6. If the mouse is anesthetized using general anesthesia, the animal should be recovered individually in a clean cage after the biopsy is completed. The mouse should be fully ambulatory before it is returned to the original cage or co-housed with other mice.

7. An alternative option for tissue sampling is the ear punch. This provides a consistent sample size between animals and does not require anesthesia. This is a less invasive means of obtaining the sample and can be performed in conjunction with ear-tag identification of individual animals (Norway, Netherlands) (2). Small quantities of blood from distal veins may be used for genotype analysis (3). Also, PCR analyses using saliva, hair, and fecal samples have been described (4-6).

References:

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3. Campbell, D.B., Hess, E.J. Rapid genotyping of mutant mice using dried blood spots for polymerase chain reaction (PCR) analysis. *Brain Research Protocols*, 1:117-123, 1997.
4. Irwin MH, Mofatt RJ, Pinkert CA. Identification of Transgenic Mice by PCR Analysis of Saliva. *Nature Biotechnology*, 14: 1146-1148, 1996.
5. Schmitteckert EM, Prokop C, Hedrich HJ. DNA Detection in Hair of Transgenic Mice – A Simple Technique Minimizing the Distress on the Animals. *Laboratory Animals*, 33(4): 385-389, 1999.
6. Broome, R.L., Feng, L., et al. Non-invasive transgenic mouse genotyping using stool analysis. *FEBS Letters* 462:159-160, 1999.
7. BVAAWF/FRAME/RSPCS/UFAW Joint Working Group on Refinement. Section 15: Tissue biopsy collection for genotyping. *Laboratory Animals* 37 (Suppl 1): S1:27-33, 2003.