Subject: Rodent Identification

BACKGROUND

Individual animal identification is important for animal colony management (genotype identification, to track breeding crosses, etc.), for animal health/medical records, and for research data. There are several methods available for identifying rodents, including ear notches, ear punches, ear tags, tattoos of the toes/feet or tail, and subcutaneous transponders. Toe-clipping should be used only when no other individual identification method is feasible. Please see the WSU IACUC Rodent Toe-Clipping Policy.

IACUC Guidelines

**Ear notch:**

1. Advantages: Simple, inexpensive, easy to read, can be done at 2 weeks of age or older without anesthesia, tissue can be used for genotyping
2. Disadvantages: If not done correctly, the tissue may grow back. May be difficult to achieve a precise and extensive numbering system.
3. Procedure:
   a. Rodent pups should be gently restrained with gloved hands by scruffing the dorsum high on the neck to control both the body and the head.
   b. A small wedge-shaped notch can be created on the outer edge of the pinna (external ear).
   c. Sterilized very sharp scissors work well for this purpose.
      i. The instrument should be sterilized prior to the first use, and cleaned with a high level disinfectant (spor-klenz, clidox, Accel) between each cage of animals.
      ii. If genotyping, the instrument should be disinfected between each animal to prevent sample contamination.
   d. Amount of tissue should be kept to a minimum. A general guideline is a small wedge of tissue not wider than 2-3mm at the ear edge.
   e. Bleeding should be immediately controlled by gentle fingertip pressure.
   f. Notches may be placed on both ears in a pattern that correlates to a numbering system.

**Ear punch:**

1. Advantages: Simple, inexpensive, easy to read, can be done at 2 weeks of age or older without anesthesia, tissue can be used for genotyping
2. Disadvantages: If not done correctly, the tissue may grow back or tissue can tear. May be difficult to achieve a precise and extensive numbering system. Punch instruments may get out of alignment causing pain and tissue damage by poor cutting action. If any drag or catching is noticed, the punch must be discarded.
3. Procedure:
   a. Rodent pups should be gently restrained with gloved hands by scruffing the dorsum high on the neck to control both the body and the head.
   b. An ear punch instrument is used to puncture the pinna (external ear). Punches may be made on the outer edge or in the middle of the pinna. Do not punch too close to the head where the cartilage is thicker and more blood vessels are present, because it is painful.
   c. The ear punch should be cleaned with a high level disinfectant (spor-klenz, clidox, Accel) before first use and then between each cage of animals.
      i. If genotyping, the instrument should be disinfected between each animal to prevent sample contamination.
d. Bleeding should be immediately controlled by gentle fingertip pressure.
e. Punches may be placed on both ears in a pattern that correlates to a numbering system.

**Ear tag:**

1. Advantages: Ear tags can be placed starting at ~2 weeks of age. The procedure does not require anesthesia. Extensive numbering system, up to 4 digits may be achieved. Ear tags can be sterilized.
2. Disadvantages: If not placed correctly, the tag will be ripped out by the rodent or fall out on its own. Numbers are very small and difficult to read. Some animals have tissue reactions to the metal resulting in crusting and thickening of the cartilage. Cannot be used in small neonates.
3. Procedure:
   a. Ear tags are placed with an ear tag instrument.
   b. Proper location is important; the middle lower pinna is used. Pictures are available in the DLAR facilities.

**Toe/foot tattoo:**

1. Advantages: Can be done at any age without anesthesia. Extensive numbering system may be achieved. Identification is permanent.
2. Disadvantages: May be technically challenging. Numbering system may be difficult to read. Ink may stain draining lymph node.
3. Procedure:
   a. Take a small gauge (25-27g) needle and dip it in tattoo ink [India ink will work] and make small dot(s) in their tails, feet and/or ears.
   b. If you just need to identify pups within a cage until they're old enough to permanently tag, this works well. Using tattoo ink of various colors enables you to make up a legend based on colors of dots, not just numbers/location of dots.

**Tail tattoo:**

1. Advantages: Can be done at any age without anesthesia. Extensive alpha-numeric system with endless possible combinations. When done properly, the tattoo is permanent. Automation is available (DLAR's Somark tail tattooing machine).
2. Disadvantages: If not placed correctly, the tattoo ink will be shed with the outer skin cells of the tail. Characters may be small and/or difficult to read. Tattooing by hand can be difficult and takes a lot of practice. Cannot be used in small neonates. Ink may stain draining lymph node.
3. Procedure:
   a. Free-hand: a tattoo machine with a fresh needle is used. Needle is replaced when dull or about every 50 animals. The needle is dipped into tattoo ink then applied with firm pressure to the surface of the tail. Tattoo oil or tincture of green may be used to enhance ink uptake.
   b. Somark® Labstamp™ tattoo machine: this machine may only be used on mice >3 weeks of age. It requires special training by DLAR before use. Briefly, the mouse is placed in a restrainer and the restrainer is inserted into the machine. Alpha-numeric identification is entered in the machine, and the machine will automatically tattoo that identification onto the tail. This process only takes a few seconds.

**Subcutaneous transponder, a.k.a. microchip:**

2. Disadvantages: Expense: microchips are expensive, chip readers are required and expensive. Recommended to be implanted at 3 weeks of age or older (mice must be sufficient size for implant). Anesthesia may be needed to facilitate implant (although not required).
3. Procedure:
   a. Sterile transponders are placed with a sterile trocar into the scruff (intra-scapular area).
   b. Test that the chip reader can detect and read the transponder.