Colony Management

Version: 1
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Summary:

Colony management encapsulates the day-to-day management of a colony. Services include breeding, weaning, identification, tissue collection, litter checks, sex differentiation, and overall record keeping of a colony.

Reagents and Materials:

Reagent/Material	Vendor	Stock Number
PPE (laboratory coat, shoes/booties and exam gloves)		
Mosaic software	Virtual Chemistry, Inc.	
Computer with internet access		
Pens (sharpie and dry erase)		
Dymo printer	Dymo	
Litter cards		
Mouse cage setup (sterile cage, food, water, enrichment)		
Cage cards		
Disinfectant		
Ear notchers	Kent Scientific	INS750078-10 INS750076-2
Small iris scissors	Fine Science Tools	14084-08
75% ethanol		
Silver nitrate, Quick Stop powder		
Isoflurane		
Ketamine		
Xylazine		

**Materials with no vendor listed may be obtained from different sources based on availability.*

Protocol:

1.0 Breeding:

Colony management staff is responsible for overseeing several different types of breeding methods, including continuous breeding and sequential breeding, in trios and pairs as approved in our protocol.

1.1 Procedure for Continuous Breeding (or Intensive Mating Systems)

1.1.1 In continuous breeding, dams and sires remain in the same cage continuously. This allows for the highest productivity for a single breeding unit. It takes advantage of postpartum estrus and because of this, the next litters will likely be born around the time of weaning the previous litter.

1.1.2 A breeding unit (a cage of breeders) can be set up with a single male and one or two females. If using two females, they will usually cycle together and be impregnated around the same time; after giving birth they will assist each other in rearing the young.

1.1.3 Method

1.1.3.1 A single male is housed with either a single female (also known as pair mating) or a pair of females (also known as trio or harem mating). The male is allowed to stay in the cage with the females for as long as needed, unless any of the mice show signs of aggression or injury.

1.1.3.2 If there have not been any litters or signs of pregnancy after 30 days, the PI or lab must be notified.

1.1.3.3 See section 2.0 below for information on weaning mice. Litters must be weaned as close to 21 days old as possible to avoid overcrowding and possible death of new litters.

1.1.3.4 Some transgenic lines have delayed weaning as pups can be very small or runted. These pups may stay with the parents until 28 days of age.

1.2 Procedure for Sequential Breeding (or Non-intensive Mating Systems)

1.2.1 This method is primarily used at MBP for generating pseudopregnant females, but can also be used for traditional breeding, especially in cases where the male exhibits aggression to his offspring. This also increases your production control as the females only have the opportunity to become pregnant when they have been set with stud males.

1.2.2 Dams are placed into cages with males and removed after a set period of time, when visibly plugged, or at the end of the week, depending on the project needs. In this method, it is permissible and indeed common to place multiple females in with the male.

1.2.3 Method

1.2.3.1 A single male is housed with either:
A single female (pair housed)
A pair of females (trio mating)

1.2.3.2 Females are removed from the breeding cage when they are either obviously plugged, pregnant, or after a set period of time depending on the project needs.

1.2.3.3 NOTE: If a female with litter is housed without a male, DO NOT place a male in the cage with her before the litter is weaned as he, or she, is more likely to destroy the existing litter.

1.3 Procedure for Rotating a Stud Male Between Multiple Female Cages

1.3.1 There may be instances where a male may be rotated between multiple female cages. Some reasons for this may include:

1.3.1.1 The mice may be shipping and offspring are desired prior to shipment.

1.3.1.2 There is a concern with how old the breeders are.

1.3.1.3 Germline transmission is desired at a quicker rate.

1.3.2 Method

1.3.2.1 The male is set up pair or trio housed with female companions and allowed to breed for up to 2 weeks. He is marked with a small colored cage card with the date he is to be rotated into a new cage of females.

1.3.2.2 On the desired date, the male is rotated into the new cage of females. A new date is written on the colored cage card as to when he is to be rotated back into the original cage.

1.3.2.3 If there is a litter in the cage, the male must not be placed in the cage with the breeder females. A new date must be written on the cage card after the wean date of the litter.

1.4 General Breeding Procedure

1.4.1 Housing of stud males

1.4.1.1 All stud males are singly housed either on one side of a duplex or in a single cage to prevent fighting.

1.4.2 Housing of pregnant and lactating females

1.4.2.1 Pregnant or lactating females can be housed singly or with another pregnant female with or without the breeding male. Breeding females will assist each other in raising their litters.

1.4.2.2 Cages with litters of pups should be marked with the date of birth of the litter.

2.0 Weaning:

2.1 As much as is practical, pups will be weaned before the next litter is born, generally between 21-23 days. For strains with smaller or runted pups, weaning up to 28 days is permitted to ensure the pup's survival (see section 2.2). Hearty mice need to be weaned as close to 21 days as possible to prevent overcrowding in breeding cages.

2.2 In the case of runted litters or litters containing 3 or fewer pups, the litter may be left with the dam(s) up to 28 days of age to allow for growth and/or aging up of companions from other litters within the colony to ensure survivability and/or social housing of weanlings. In the case of multiple litters where pup crowding/competition may be a concern, older pups with fully developed proprioception and higher stress potential will take priority over newborns and will remain in the cage until they are of appropriate wean age/size.

2.3 As approved in our protocol, mice can be bred in trios and will remain housed in the same cage through pregnancy and weaning of litters. Up to two pregnant females, the male, and litters may be housed in the same cage.

2.4 Weaned pups must be separated by sex and housed up to 5 mice to a wean cage.

3.0 Identification:

3.1 Rodents may be individually identified in addition to having an appropriately labeled cage card.

3.2 Appropriate identification for mice includes ear tag, ear notch, tail/toe tattoo, microchip, toe clipping.

4.0 Tissue Collection:

4.1 Some projects may require genotyping while housing mice at UCD. Genotyping sampling may be done by tail snips, ear punches, or toe clipping, depending on age. Altricial pups will be sampled by "toeing".

4.2 Tail Clipping

This method involves amputating a very small segment of the distal tail. At <17 days of age, the degree of ossification of the coccygeal vertebrae in the distal 5 mm is much less than that at 1 cm. After 17 days of age, the degree of ossification is similar at the distal 5 mm and 1 cm tail segments. Perception of pain is assumed to be more likely in bony versus cartilaginous tissue. Tail clipping on mice or rats ≤17 days of age does not require anesthesia. Animals must be appropriately restrained during the procedure to minimize trauma. Sterile sharp scissors (must be disinfected between uses) or a sterile blade per animal can be used for the procedure. Only the distal 2-5 mm should be amputated. Hemostasis can be achieved by using a silver nitrate stick, Quick Stop powder, or by applying a gauze sponge over the site with gentle pressure until bleeding stops. Animals should be observed closely after returning them to their cage to ensure hemostasis. CVS must be contacted if the lesion shows necrosis, bone exposure, and/or the animal's activity level is not normal.

Age	Anesthesia	Analgesia	In Protocol
≤ to 17 days	Not necessary	Not necessary	Yes
>17 days or animals requiring more than one tail sample	Yes	Yes	Yes

Animals >17 days of age that require tail clipping must be under general anesthesia using ketamine/xylazine or isoflurane during the procedures and administered a systemic analgesic (i.e., buprenorphine, meloxicam, carprofen) given at least once prior to the procedure. The use of these medications must be clearly detailed within the procedures outline of the approved protocol, including the drug table. **This method is per an approved protocol. If multiple tail clippings are required a maximum of 1 cm total tail length can be amputated, with all tail clippings combined."

4.3 Ear notch/punch

This method involves punching a hole or making a notch in the ear pinna. Commercial ear punches are available and inexpensive. Ear notching using an ear punch is a permanent form of identification. Ear notch remnants can usually provide enough tissue for DNA sampling during the initial PCR screening. Ear punch samples collected on animals do not require the use of anesthesia or analgesics, however, for identification purposes the animal must be appropriately restrained to ensure proper technique. The ear punch device used must be disinfected between cages of animals. These devices can be autoclaved.

4.4 Toe Clipping This method involves removal of the distal phalangeal (coffin) bone of one or more limbs. Toe clipping, as a method of identification of small rodents, should be used only when no other individual

identification method is feasible and should be performed only on altricial neonates (≤12 days). Toe clipping has the potential to induce pain and distress, alter the animal's gait and ability to feed. Mice older than 12 days of age must be anesthetized for this procedure to ensure proper technique and to minimize pain and distress. Only one toe per foot may be removed**. A pair of sterile sharp scissors can be used for this procedure (must be disinfected in between uses). Hemostasis can be achieved by using a silver nitrate stick, Quick Stop powder, or by placing a gauze square over the site and applying gentle pressure until bleeding has stopped. ****Per the approved protocol for MBP colony management, some pups may have two toes per foot clipped.**

5.0 Euthanasia:

5.1 Please refer to the UC Davis- Conventional husbandry protocol.