

Quick Reference: Improving Mouse Breeding Success

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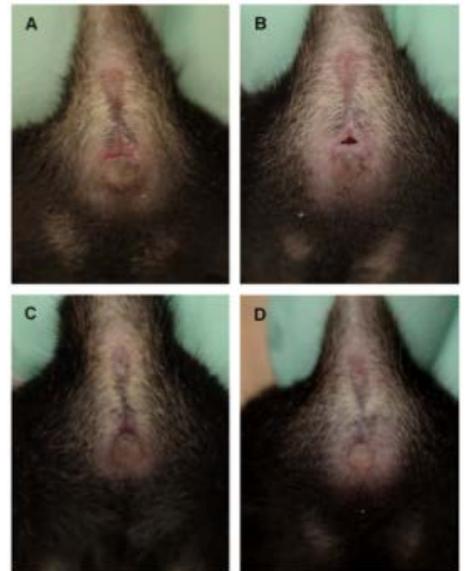
Environmental Factors:

- Researchers should keep the amount of noise (audible and ultrasonic) and traffic in breeding areas to a minimum. Handle cages calmly and quietly.
- Place cages low on racks and in positions on racks that are farther from housing room doors and cage change stations.
- Preservation of the standard light dark cycle in the housing room is critical (see Dauchy et al). Common sources of light at night contamination include cage changing stations, biosafety cabinets, and hallway lights via either an open door or window.
- Enrichment: Appropriate nesting material (e.g., Bed-r'Nest® puck) improves breeding and pup survival (see Gaskill et al).



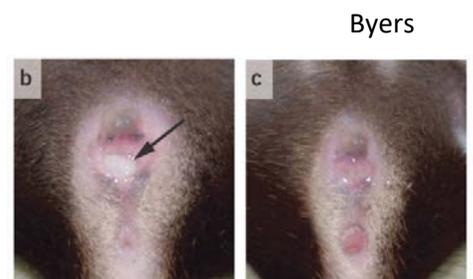
Mating:

- Use females at least 6 weeks of age and male mice at least 8 weeks of age.
- Replace older breeders with younger mice on a regular schedule. Replace females at 8 months (6 months for many transgenic lines). Replace males 1 year of age (8 months for some transgenic lines).
- Replace females with a history of cannibalizing litters, failing to produce litters in 3 successive breedings, or producing litters much smaller than the strain is expected should be replaced.
- Females in group housing may go into anestrus. Male pheromones from dirty bedding of a male's cage stimulates estrus in females (Whitten Effect).
- Allow male mice at least one day to get used to a new cage prior to beginning mating.
- Check for estrus: estrus lasts 15 hours with ovulation occurring in the middle of the dark cycle.



A. proestrus (receptive to mating)
B. estrus (receptive to mating)
C. metestrus
D. diestrus

- Females which appear swollen, pink, and moist in the late afternoon is receptive for mating at night.
- Mate mice overnight and confirm presence of vaginal plug. Note that plugs from B6 mice may be thin and dissolve quickly.
- When handling multiple breeding cages, scent marks present from the previous cage must be removed prior to handling the animals in the next cage. Practice strict microisolator technique and keep gloved hands wet with disinfectant. Exposure of a recently bred female to the pheromones of a strange male can lead to pregnancy loss (also known as the Bruce Effect).
- Cycle females through a male's cage to maximize production from a small number of males. Do not cycle males through a female's cage.
- Homozygous mice that are severely impaired (infertile, embryonic lethality, die in utero, or die before reaching sexual maturity) can be backcrossed. Progeny must be genotyped.



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- Inbred mutants should be back-crossed to the appropriate parental inbred strain every 10 generations to prevent genetic drift. Mutants on a mixed genetic background can be backcrossed to appropriate F1 hybrids about every 10 generations

Gestation and Post-Parturition:

- Do not disturb gestating females more than necessary
- Avoid changing cages with new litters for 3 days after birth. Indicate a request for delayed cage change with the cage card provided by OLAC. At the end of the 3 days, the lab must change the cage. If moving pups aged P0-P7, scoop the entire nest up with a gloved hand to transfer pups with minimal disturbance. If individual pups must be handled, spray gloved hands with appropriate disinfectant prior to handling.
- Investigators must record the birth and/or weaning dates on the appropriate breeding card.
- If dead/cannibalized pups are observed, check remaining pups for milk visible in their stomachs (i.e., a milk spot). If pups are not nursing, cross foster or euthanize pups to prevent starvation.
- Prior to weaning, certain strains may benefit from offering moistened food on the floor of the cage. Runted pups may also be offered dietary supplements (e.g., DietGel® Boost, Nutri-Cal®, etc.).



Weaning:

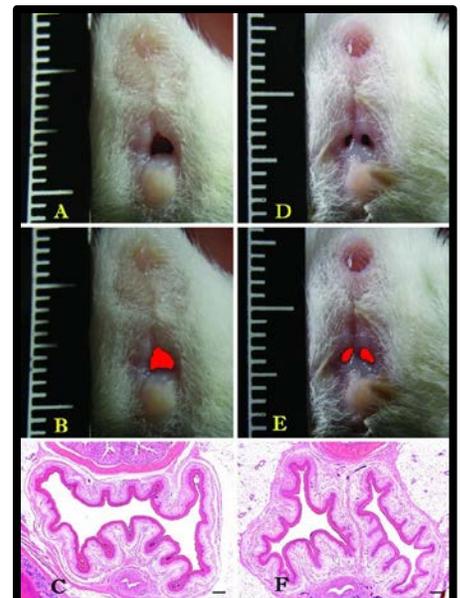
- Provide moistened food on the floor of the cage of all new weanlings, in addition to standard diet in the food hopper. Replace moistened food daily. Supportive care post-weaning may need to be extended for individually housed weanlings (e.g., single male in litter).
- Pre shred nesting enrichment for small weanlings to provide thermoregulation until pups are able to nest on their own.
- If trio breeding is approved in your AUP, litters may require weaning prior to P21 (e.g., P19-20) to avoid disparate litters and newborn pup loss. In such cases, provide supportive care post-weaning.
- If utilizing post-partum estrus, pups need to be weaned prior to P21 (e.g., P19-20) to avoid disparate litters and subsequent newborn pup loss.
- Some transgenic strains that are prone to runt pups may require delayed weaning. This must be approved in your AUP for specific strains. Trio breeding and the use of post-partum estrus is discouraged to avoid production of disparate litters.

Breeder Diet:

- Breeder Diet should be avoided in male mice, as obesity inhibits breeding. Breeder Diet (diets high in fat) should only be used when females are present.

Health conditions:

- Decreased or increased BCS. Animals that have a decreased body condition, should not be bred. Reproduction in obese mice is decreased.
- Dystocia is defined as abnormal labor or birth. This difficulty may be due to a number of different factors. Dystocia that has been determined to be the result of genetic abnormalities should prompt euthanasia of the dam. Causes include:
 - Uterine inertia
 - Exhaustion/energy deprivation
 - Deformed pups
 - Vaginal septum (see Chang et al, Figures D/E)



Additional References

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