

Guidelines for the Establishment and Use of Mouse Breeding Groups

Background: This guideline is intended to define considerations for the establishment and maintenance of mouse breeding groups consisting of one male and up to four females who are reproductively viable (1). These guidelines are not intended to address breeding groups with more than four females since these breeding schemes will be addressed by individual Institutes/Centers (ICs).

Pair breeding (defined as one adult male and one adult female) will minimize the chance of overcrowding, simplify colony management, and allow identification of the dam with litter. Trio breeding groups (defined as one adult male and two adult females) have been used successfully at the NIH and other institutions for several decades, especially for strains that are difficult to propagate. The use of continuous trio breeding groups can result in a higher breeding efficiency by permitting breeding at the first post-partum estrus and facilitating pup survival through cross fostering. The Jackson Laboratory recommends mouse trio breeding groups, stating that, "Most strains produce more progeny per cage if mated as trios because all adult cage mates generally help care for the young." (2) Harem mating (defined as one adult male and three or four adult females) maximizes the progeny of an individual male but may sacrifice the first post-partum estrus if pregnant females need to be removed from the cage to avoid overcrowding. Harem mating may also result in injury to the breeding male. Harem mating is an option for projects that require an intensive breeding program.

The discussion of rodent cage size and density in the *Guide for the Care and Use of Laboratory Animals (Guide)* (3) makes it clear that institutions should use those space recommendations as a starting point for determining caging requirements. Furthermore, the *Guide* identifies examples of performance indices to assess the adequacy of housing which include the animal(s) health, reproduction, growth, behavior, activity, and use of the space. Both the NIH Office of Laboratory Animal Welfare (OLAW) (4) and AAALAC International (5) have stated that mouse cages of the size commonly used in the United States may be appropriate for trio breeding groups, with the caveat that there are several factors that need to be considered when assessing the adequacy of cage space. For example, a mouse litter size can vary greatly depending on the age of the female or whether the strain is inbred or outbred. The *Guide* (3) recommends that sufficient space is available for mothers with litters to allow pups to develop to an appropriate weaning size and weight without detrimental effects for the mother or litter. Each program/facility's Animal Care and Use Committee (ACUC) is responsible for the review and approval of a policy for mouse breeding groups that considers a range of factors which may impact the appropriate density including but not limited to the style of cage, the types of bedding/enrichment in use, cage change frequency (6-11), the number of adults, number of pups, and age of various litters (12) to ensure the adequacy of cage space. In shared animal facilities, multiple IC's policies may be in play; ultimately it is up to the lead IC to determine which policies can be supported.

Investigators who decide that trio breeding groups or harem mating would be a beneficial breeding strategy for their research must accept responsibility for any overcrowding in their colony, and follow these guidelines:

1. Trio breeding groups are best suited for the propagation of inbred, transgenic, or other strains

of mice which generate small numbers of pups or are difficult to breed. Crosses that produce larger litters are best propagated by a pair (defined as one male and one female) breeding strategy.

2. Investigators using a breeding strategy that includes harem or trio breeding groups must be aware that they have increased responsibilities to ensure “the adequacy of cage space” for all animals (13). These increased responsibilities include the prompt weaning of mature pups between 18 and 28 days of age and the prompt separation of animals if the cage density becomes greater than that established by the facility or program. In addition, difference in the age of the pups of different litters, growth rate, cage dimensions, and husbandry practices such as cage sanitation (14), adequacy of preventing fecal contamination of food/water/nesting materials and ensuring these resources are accessible to all cage occupants may require an investigator’s prompt attention and intervention. Many programs separate litters in the same cage that are born more than 7-10 days apart, with a mother or their respective mother, to prevent the older pups from injuring the younger pups.

References:

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