

RATS

DATE OF PUBLICATION: September 2022

These frequently asked questions (FAQs) are intended to assist investigators, instructors, and members of animal care committees in the implementation of the [CCAC guidelines: Rats](#) (CCAC, 2020). FAQs provide general responses to comments and questions received by the CCAC during the external reviews of this guidelines document.

If you do not find the answer to your question here, do not hesitate to [contact the CCAC](#) and we will be pleased to provide assistance.

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1. Why is there no table of cage size dimensions provided?

The guidelines document takes a performance-based approach for cage size, as stated in Guideline 2: “Cages should be of a sufficient size to allow rats to be housed in appropriately sized groups and to perform behaviours important to their welfare.” General engineering standards for all rats are difficult to set as rats of different strains or rats that have undergone genetic modification may have different requirements for the environment in which they live. Some minimum standards are provided, with examples of how to meet the behavioural needs of the animals. In addition, the rapid growth rate of rats must be considered in relation to the length of time that they will be held and the potential for their needs to change in that time period. The reproductive status of the rats also needs to be considered, including the weaning size/weight of all pups, and the space requirements for all adult animals that are present. Appendix 2 of the guidelines document emphasizes the importance of group housing, as rats are a highly social species, and lists other elements important in assessing the suitability of the cages.

2. Why is cage height important?

The ability to stretch vertically has been shown to be an important behaviour for rats, which should be accommodated by providing sufficient height in at least a portion of the cage (Büttner, 1993; Makowska and Weary, 2016). There is no prescriptive dimension provided for cage height, as it needs to be assessed for the individual rats, being highly dependent on the age and size of the rats.

3. For metabolic cages, the requirement for a resting platform and the means for rats to establish a microclimate when they are held for more than 24 hours would interfere with measurements (e.g., urine collection) and possibly negate the use of metabolic cages—how can this be addressed?

As described in the guidelines document, there are several aspects of metabolic cages that can have a negative impact on the welfare of rats, including the requirement for single housing, the barren environment, and the use of perforated or wire flooring. The 24-hour timeframe is motivation for designing studies that do not require rats to be kept in metabolic cages for more than 24 hours, or including innovative collection methods that accommodate the presence of platforms and other resources (e.g., extracting urine from paper towel). Providing enrichment can improve the welfare of rats in metabolic cages; however, they will still be negatively impacted by the lack of social interaction, the barren environment, and limited ability to engage in coprophagy.

4. Are both bedding material and nesting material required for rats? What types of material should be used for each?

Both bedding and nesting material are important to enable rats to create a comfortable microenvironment and allow them to perform some natural behaviours, such as digging, burrowing, and foraging. However, the specific composition of these two types of materials allows them to serve different functions. Bedding material needs to be able to absorb moisture from urine and feces and control ammonia levels, while nesting material should be easily manipulated by rats for the construction of nests. Bedding material is not needed for thermoregulation if nesting material is present. The extent of nest building by rats has been shown to vary with strain (Jegstrup et al., 2005) and early exposure to nesting materials. In general, rats prefer paper strips that are easy to manipulate (Manser et al., 1998).

5. How important are shelters for rats?

Shelters should not be considered to be environmental enrichment; rather, they should be standard components of rat housing that address a variety of activities that are part of the normal rat behavioural repertoire. These include withdrawal from light, control of microclimate (which aids in thermoregulation), escape from aggressive social interactions, thigmotactic avoidance of open spaces, nest-building activity, and gnawing (depending on the materials that structures are made from). As they are a social species, rats seem to prefer shelters with a roof and three walls that will accommodate at least two adults at once.

6. Can rats be singly housed?

Single housing of rats should only be permitted when there is strong scientific, welfare, or medical justification, as rats are highly social. If it is necessary to singly house rats, it should be for the shortest duration possible, and visual, auditory, and olfactory contact should be provided. A cage divider with “social holes” is one way of providing contact when it is necessary to separate rats for individual measurements.

7. Is toe clipping permitted as a method of identification?

Toe clipping is a highly invasive procedure and is not permitted solely for the purpose of identification. It is only permitted when no other individual identification method is feasible and the tissue collected is also needed for genotyping or other protocol requirements. Justification for using toe clipping as a means of genotyping, rather than less invasive methods, needs to be provided to an animal care committee. Only one toe is to be clipped.

8. What basic needs of rats must be addressed, and what forms of environmental enrichment can provide added welfare benefits?

The guidelines state that the basic requirements for rats are group housing, shelters, and sufficient space to accommodate these elements and behaviours important to their welfare.

As stated in the [*CCAC guidelines: Husbandry of animals in science*](#) (CCAC, 2017), “Measures should be sought to improve the welfare of the animals by enriching their environment in a manner that further addresses their species-specific and individual physical and behavioural needs.” In determining appropriate forms of enrichment, consideration must be given to the individual animal and the research requirements, including biosecurity issues, and any enrichment provided should be evaluated to ensure it is providing the intended benefit. Some potential forms of enrichment for rats are listed in the guidelines document, and include features to promote activity, substrates for burrowing, structures to facilitate hiding or avoidance, objects to chew, variety in food, and foraging opportunities.

9. CCAC guidelines documents now put greater emphasis on welfare assessment. What is expected and what tools are available to assist in welfare assessment?

Welfare assessment has always been a necessary component of animal-based studies, both for ensuring a good quality of life for the animals within the constraints of the study, and for the quality of scientific data. In recent years, there has been more emphasis on this in the literature, and CCAC guidelines documents

have responded by drawing more attention to this important aspect of animal-based science and highlighting the development of tools that can inform welfare assessment.

As noted in the [CCAC guidelines: Rats](#) (CCAC, 2020) the investigator, in consultation with the veterinarian, is responsible for planning and documenting welfare assessment; however, the collection of information should be integrated with the activities of all personnel involved with the animals for more efficient use of resources and less disturbance to the animals. The overall focus of any welfare measurement should be to identify the need for mitigation strategies or endpoints, with the aim of improving the welfare of the animals (i.e., monitoring should be conducted to benefit the animals, not for the sake of monitoring).

Section 8, “Welfare Assessment”, in conjunction with Appendix 4, “Indicators that May be Used to Assess the Welfare of Rats”, provide a number of references to help investigators and animal care committees identify useful indicators.

10. Since there is evidence that isoflurane is aversive to rats, is it still recommended prior to using carbon dioxide for euthanasia?

The [CCAC guidelines: Rats](#) (CCAC, 2020) note that exposing rats to carbon dioxide for euthanasia is likely to cause pain and distress (Leach et al., 2004; Moody et al., 2014). The guidelines document further states that inhalant anesthetics have also been found to be aversive to rodents, but that isoflurane has been shown to be less aversive to rats than CO₂ (Makowska et al., 2009). However, more recent publications show that aversion to isoflurane can increase with repeated exposure (Bertolus et al., 2015; Wong et al., 2013). Isoflurane is recommended for use if it is the animal’s first exposure to it.

The guidelines document acknowledges that there is currently a substantial amount of research being conducted on inhalant techniques for euthanasia, and encourages careful evaluation of any new evidence that becomes available. Recent papers in this area are cited in the [CCAC guidelines: Rats](#) (CCAC, 2020) (Valentim et al., 2015; Baker and Hickman, 2018).

11. What are the implementation timelines for the new housing requirements?

The CCAC has determined that all existing, non-compliant rat cages within certified institutions must be replaced by the following schedule: 33% by April 21, 2025, 66% by April 21, 2028, and 100% by April 21, 2031. This decision is based on fairness, consistency, animal welfare considerations, and current supply of larger cages. Please note that all newly acquired rat cages must meet the housing guidelines described in the [CCAC guidelines: Rats](#) (2020). Moving forward, the CCAC will actively seek updates from program participants on their progress toward these goals during assessment visits and communications.