

# *environmental enrichment*



# *institutional standards*

*Does your facility have standards implemented for rodent environmental enrichment?*



- All our mice get tissue paper and cardboard—usually toilet paper rolls or cut up glove boxes and egg cartons—for nest building and shelter. We also provide chalk sticks for gnawing.
- The cages of our mice are each furnished with a commercial plastic house, a running wheel, and bedding material such as a cotton fiber pad, paper towel or shredded paper.
- The standard furniture of our rat cages consists of one plastic tube serving as refuge and a piece of wood for gnawing.
- We have made it a standard rule that every single-housed rodent (a) has access to a toy that is rotated once a week, and (b) receives daily produce and grain.

When submitting an IACUC protocol, there is a question if the individual animal

can receive enrichment or not. If the answer is no, there has to be scientific justification. So far, no investigator has ever insisted that the toy be removed, but some have placed restrictions on food enrichment.

- At our facility:
  - (a) the mice get nesting material and a commercial plastic house,
  - (b) the rats get a plastic tube and a hardwood block, and
  - (c) the guinea pigs get a plastic house, a hardwood block and autoclaved hay.

These enrichment provisions are standard for every animal. If researchers want to opt out, they must provide a written explanation in their protocol; it has to be a very good reason.

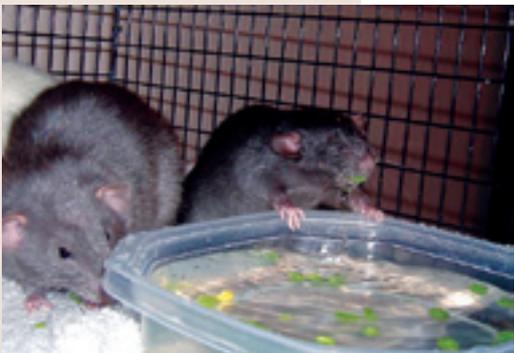
We encourage everybody working with our rodents to offer food treats as positive reinforcement after procedures.

# foraging enrichment



*What are practical options to promote foraging activities in rodents?*

- Giving rats large seeds by hand certainly prompts much hoarding. I also like to supply them with whole walnuts that they gradually learn how to open. They enthusiastically pull scrunched-up paper out of toilet paper rolls, chew through little cardboard boxes and dig through a pile of shredded paper to retrieve hidden treats. Frozen peas floating in a shallow pan of water is another popular attraction for them.



- For rats and mice, I place high-fiber rabbit food in the bottom of the cage at cage changing; this provides for low calorie, inexpensive foraging enrichment. Hay is also great, both as food enrichment and nesting material.
- Our hamsters receive irradiated sunflower seeds, dried fruit, and peanuts.
- At my institution, the breeding groups of guinea pigs get fresh green grass, vitamin C dissolved in water, and autoclaved hay.
- Based on my two-year experience with a large breeding group of guinea pigs, I can say that guinea pigs relish dandelions and other thoroughly washed green stuff from the garden; they love high-quality hay—the more the better—without experiencing adverse side effects.



- Our guinea pigs relish lettuce, carrots and apples. We've tried cabbage, but some animals don't tolerate it too well, so we stopped feeding it.
- Fresh green stuff is a disease risk, as it can be contaminated; bird droppings are a particular problem. Washing the material in clean running water will remove much of the contamination but it's not foolproof. Many years ago, we fed cabbage and kale to guinea pigs and lost a major colony due to salmonella-contaminated kale that had been washed but obviously not thoroughly enough. We do feed raisins and other dried fruits of human consumption quality to all our rodents.



# investigators' permission

*At your institutions, do you have to ask permission from the principal investigators (PIs) to enrich the cages of rodents?*

- We have a form that all PIs sign, giving permission for enrichment. As more and more investigators become educated about the value of enrichment [for example: Cooper & Zubek, 1958; Diamond, et al., 1964; Bennett et al., 1969; Ferchmin et al., 1970; Carughi et al., 1989; Fernandez-Teruel et al., 2002; Arendash et al., 2004; Cancedda et al., 2004; Green et al., 2004; Neugebauer et al., 2004; Van de Weerd et al., 2004; Sharp et al., 2005; Fox et al., 2006], some actually ask us to provide more enrichment for their animals; this is so nice to see! However, our IACUC will give researchers permission to remove enrichment if they have a convincing reason for it.
- At our facility, it is explained to the researcher that each animal cage has to contain certain enrichment objects or structures. An investigator has to have compelling reasons to keep his/her animals without such enrichment. When we get to know the techs working with the animals, we encourage them to distribute food treats, like cereals and also autoclaved hay, if this does not interfere with the research.



I find that the techs are very willing to give these items to the animals in their charge. It makes all involved feel much better!

- Before adding any kind of enrichment in the cages, we first seek the permission from the investigator to make sure that the enrichment does not affect the experiment. Our investigators have to inform us proactively if there are certain enrichments that must be withheld for study reasons.
- In our facilities, we have begun the process of outlining in our IACUC protocol application forms what we consider to be standard practice, such as provision of nesting materials and shelters for mice,

rats and hamsters. Researchers are asked to specify what enrichment provisions they want to be withheld and explain exactly how these enrichments would confound their experiments.

I do see an increased interest and willingness in many of our researchers to provide their rodents an optimal environment, which I attest to a better understanding of the effects of improper environments on research data. There's still much room for improvement, but I think we are moving in the right direction.

- It is my impression that provision of basic environmental features is still not routine in the United States. The implementation of environmental enrichment in a facility seems to depend heavily on the motivation of key people.
- I agree, environmental enrichment is often left up to the individual caretaker; there is no policy that has to be followed. The problem with this is that some animal care staff believe greatly in enrichment while others think it is unnecessary. This inconsistency is probably natural but it is very frustrating; without institutional standards, some animals will have the benefit of having their living quarters enriched by motivated personnel while others have to be content with barren cages because the attending personnel are not interested in providing environmental enrichment.

- At our institution in the United Kingdom, environmental enrichment is the norm. If researchers want to opt out, they must include scientific justification in their project license application so that it can be taken into account by both our internal ethical review process and the Home Office when they consider the license for approval.
- Environmental enrichment is something that Brazilian researchers are not aware of at all. People usually do not give any environmental enrichment for rodents or rabbits, especially if it is a short-term experiment. Therefore, the question of obtaining permission to provide the animals with enrichment does not arise.





## rats



## amazing social creatures

*Is it true that rats are reliably tolerant of each other?*

- That rats get along with one another so well is one of the reasons I like working with them. I have never noticed any signs of aggression among rats.

I have worked with rats in neuropathic research. The animals were always very friendly to each other, even when they were experiencing pain. I witnessed rather affectionate interactions—such as grooming and bringing food pellets nearby—in rats who had undergone surgical procedures.

- We always keep individual rats during the post-operative phase with a buddy to speed up the recovery.
- I have worked with rats for several years but never witnessed that they were aggressive with each other. Yes, they can be aggressive with humans; I have been bitten on several occasions.

- The only time I've ever seen aggression among rats was when someone inadvertently put ex-breeder males together; predictably, this resulted in a major scrap. Unfortunately, this means that once a stud male has been used for breeding, he has to be housed singly from thereon unless he lives in a permanent pair or harem.

We occasionally get a female who becomes aggressive during the time she has a litter, but that's toward humans, not other rats.

Diabetic rats can be aggressive toward each other if their insulin is out of control, but that is only temporary and, once insulin is back under control, they become their



usual sunny selves. I believe that obese Zucker rats can be bad-tempered but suspect this is also due to erratic blood sugar levels.



- During my undergrad training, I worked in a pet store for several years. We would routinely put together rats of various ages and genders. I don't recall that we encountered any aggression-related problems when the animals met each other for the first time. To make it even more interesting, we would often use nursing rat moms to foster pups from other rodents, such as hamsters and gerbils; we simply put the new pups in the pile, and the rat mom would take care of them.
- Apart from the jumpy strains, rats are remarkably docile. They can make great children's pets.



- I have always found rats to be the most accommodating of the lab critters: well-mannered, well-groomed, social, affectionate, and intelligent. When it comes to their relations toward each other, I've only ever seen one squabble in 13 years. This occurred when an older female's roommate died and I had a singly-housed girl, who I thought would make a nice friend for her. When I placed them together,

the older female did a little song and dance number with a bit of a hiss; but I think that was just to explain that she was Queen Bee. The next day I found them sleeping in their little snuggle patch, as rats do, and they continued to have a happy relationship.

- We humans could learn something from rats in regard to living together rather than fighting against each other.

## are rats animals?

Considering the fact that rats are such amazing animals, isn't it outrageous that they—along with mice and birds—are explicitly not covered by U.S. animal welfare regulations (United States Department of Agriculture, 1989)?

- Here is the most recent update of these regulations, including the definition of the term animal:  
“Animal means any live or dead dog, cat, nonhuman primate, guinea pig, hamster, rabbit, or any other warm-blooded animal, which is being used, or is intended for use for research, teaching, testing, experimentation, or exhibition purposes, or as a pet. This term excludes: Birds, rats of the genus *Rattus* and mice of the genus *Mus* bred for use in research (United States Department of Agriculture, 2002a).”

This language creates a rather bleak situation for the majority of animals used in research, testing and education in the United States.



- Many countries have animal welfare laws and regulations. To my knowledge, the situation in the U.S. is an exception: no other country excludes any species in its legal definition of the term animal.
- Why are rats, mice and birds excluded from regulations and oversight? It's a purely economic decision. American research runs on rats and mice. And if birds were regulated, where would the research on chickens, chicken embryos, and pigeons be? In America, our laws are made by the most powerful lobbyists, not by the Congressmen themselves. In terms of animal suffering, we Americans are a lot less civilized than many of our coworkers in Europe.
- I think it is important to add that although the USDA [United States Department of Agriculture] animal welfare regulations do not include mice, rats or birds, the Public Health Service Policy on Humane Care and Use of Laboratory Animals [Public Health Service (PHS), 1996] covers all animals used in research; this includes mice, rats and birds, as well as cold-blooded animals. This policy must be followed by any institution receiving federal funds.

Additionally, many institutions—including private industry groups—are also AAALAC [Association for Assessment and Accreditation of Laboratory Animal Care International] accredited, which uses the the *Guide* (Guide for the Care and Use of Laboratory Animals) as the current standard of care. So, while there are some facilities that do not fall into either of these categories (and there are a few, I will concede), the majority of animal research does follow a standard of care similar to that described in the animal welfare regulations that are promulgated by USDA.

- As a side note, it needs to be remembered that:
  - (a) “Animal facilities should [not “must”] be operated in accord with” the Guide and the PHS Policy (National Research Council, 1996, page 2), and
  - (b) the NIH [National Institutes of Health], which administers the federal funds, is not an enforcement agency; therefore, the public has no guarantee that the Public Health Service Policy is actually followed by institutions receiving NIH grants.

## petting

Rats seem to like it when you pet/groom them. Is there a body area where they prefer to be groomed?



- I used to be a teaching assistant for a rat lab in college, and ended up *pardoning* a couple of rats who became my pets. They always seemed to be soothed by me petting them, very gently and calmly, right at the base of their neck, especially while they were falling asleep. Prior to working with them, I didn't appreciate how very affectionate they could be!
- Rats seem to enjoy having their heads gently scratched behind the ears, and some will lie down for their abdomens to be scratched. If in free mode having an out-of-cage wander on the bench, rats will come up to where someone is working and get in the way until you give in and groom the critter. I find that all of our rats—not just pets—appear to welcome a gentle scratch whenever there is an opportunity. I always encourage investigators to spend some acclimatization time with their rats before they start experimenting with them. This should make them appreciate how amazing their research subjects are and, hopefully, re-evaluate the implications of their planned experiments on them.



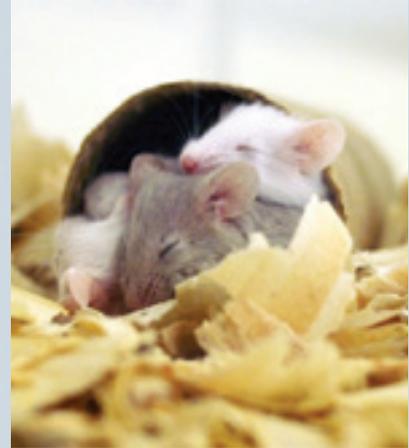
- Yes, having the investigators gently handle and groom their rats before starting an experiment is a great idea. An investigator, who has experienced the natural affection of rats and their spontaneous trust in humans, will be very careful not to cause them any avoidable discomfort or harm when conducting an experiment with them.
- If rats have been properly socialized, they will often solicit attention. They seem to love it when you gently scratch them behind the ears, on the head between the ears, and when you give them a mini-massage on the neck. I've had some rats who liked the tops of their front legs massaged too. Males are more likely to sit still for petting/grooming than females.

I love those little guys! They are really underrated creatures—extremely affectionate and playful, and great learners.
- The rats I have had as pets, and those I have worked with for a long time at work, particularly love to be petted on the top of the head and between the ears. They also give the impression that they really like it when their tails are being stroked from base to tip.
- My pet rats solicit both grooming and play, and they also groom my hands. There are studies showing that rats emit ultrasounds, a kind of *rat laughter*, when they are groomed by hand at the nape of the neck. Rats groom in this manner when they want to play with another rat [Burgdorf & Panksepp, 2001; Panksepp, 2007].
- My rats have always loved grooming me, but they've all been different. I had one girl who would grab my nose with both of her front paws and just lick away. Others liked grooming my eyelashes. Many of them preferred to lick my front teeth—I know, this is a bit gross, but for some reason it didn't bother me, probably because they were my pets. Some licked like crazy, some also nibbled a little bit with their teeth as they would when grooming their own fur. These were very special experiences for me.

- Rats are highly developed social creatures. That they typically establish such affectionate relationships with their human caregivers makes it even less acceptable that U.S. animal welfare regulations do not recognize them as animals.
- I completely agree. I once worked at a zoo facility that bred rats but kept the animals under very poor husbandry conditions. I fought to get better litter for them—they were kept on pine shavings, which are toxic to rats—and suggested that they need bigger cages. When USDA officers would come and do site inspections, they completely ignored the rat room! How can these wonderful, intelligent little creatures be so disregarded and treated as if they were disposable?
- It's so wonderful to witness such enthusiasm for rats! Rats were my first love, but sadly I became severely allergic in graduate school. This is how I ended up with monkeys. I was so accustomed to working with highly intelligent/social creatures that my heart broke when I was told that I would have to find another way to spend my career if I wanted to continue to work with animals. I, of course, love working with monkeys. Luckily, over the years, I have toughened up my immunity so that I can, once again, play with rats. Yeah, I still get hives along my neck when they ride on my shoulder, but it's totally worth it for both parties involved!
- All rat strains I have encountered (Sprague-Dawley, Fischer, Long Evans, Wistar) have enjoyed the scratch behind the ear, but I've only had Long Evans seek/allow their bellies to be rubbed.
- My rats have always loved being scratched/rubbed behind their ears and along the top of the head. Also, a few of them enjoy having their chins and/or upper backs rubbed as well. Then, of course, some will roll over for a good belly rub.
- I need to add another strain of rats to the belly rub list. We had two nude rats arrive at our facility this week, and I was assisting the research group getting used to the proper handling of these new animals. Both of the little guys were more than glad to curl up in my hands for a scratch behind the ear and then wiggled themselves into a position for a belly rub.



mice



## signs of well-being

*What are the signs warning you that a mouse, who is neither under the influence of a test drug nor of an experimental procedure, does not feel well? Are these signs the same for rats?*

- When I worked with lots and lots of mice, I looked for hunched back, ruffled—or poorly groomed—hair coat as warning signals.
- Aside from the obvious lack of food consumption or lack of urine/feces in a cage, I pay attention to the postures and movements of the mice. Typically their movements are a dead giveaway when they don't feel well. They might be sluggish, or hunched, or their gait might be a bit off.

I also check if they are grooming or interacting with their cagemates. It can take a bit of practice and patience but, once you know the normal behaviors of the animals in your care, you can usually



pick up even subtle changes in those behaviors very quickly. Since mice—and rats—tend to be good about hiding their symptoms, this can mean the difference between an easy recovery or a rapid race to the euthanasia chamber.

Unfortunately, none of those signs that I use are well-defined. It's more of the old *I know it when I see it* than look for X, Y, and Z. Obviously, we still look for appetite, weight loss, hair coat, shivering, etc., but in my experience, those symptoms tend to manifest *after* behavioral or movement changes.

- Every veterinarian will endorse your statement: the real knowing does not come from the mind. Yes, this is not a scientific approach to the question at hand, but it has proven over and over again that you often *know without thinking* that something is not right with an animal, that the animal needs help.
- Changes in posture in particular, but also piloerection, are reliable indicators that a mouse does not feel well.

- Isolation from the group, hunched posture and starry coat—hair clumping or standing on end—are, in my experience, reliable signs that a mouse is going downhill. Such an animal *feels* light, compared to a healthy animal, long before you can actually see that the animal is losing weight. Yes, you need to know your charges very well in order to recognize such warning signals before it is too late.

Introducing a favorite food, such as a few sunflower seeds or some forage mix, is also a good well-being test: if a mouse doesn't try and grab her share, then you may well have a problem at hand.

Sometimes, you can pick up a peculiar smell in a rat room that tells you that an animal is not well. This peculiar smell is more difficult to detect in mice as they have such a strong odor of their own.

When I was a young tech, listening to my animals was a good aid to detecting ill-health, as I could hear wheezing/sneezing; this was often the earliest indication of an outbreak of Chronic Respiratory Disease [CRD] and appeared long before any visible signs of the problem.

- We recently put Huntington's mice into enriched cages that contained a climbing rope and a beam. By monitoring the use of these enrichment structures, we noticed a decrease in usage by individual mice several days before any clinical symptoms of disease could be observed, and many days before they actually got sick.
- This is obviously a good way of testing for neurological diseases, without the need to disturb the mice [Carter et al., 2001; Cummings et al., 2007; Quinn et al., 2007].
- We use the ability to run/walk on a beam or cling to a rotating rod as an indication of Prion disease development in mice. The inability to perform these tests and/or the time it takes a mouse to clear food out of a tube reliably shows that she has Prion disease well before any other signs are seen; this has enabled our researchers to refine the end point of the disease dramatically.

## petting

*It seems to me that mice –unlike rats–do not like to be touched, let alone petted by a human; is that correct?*

- Mice definitely are less interested in being petted than rats, but I have found a few who tolerated, perhaps even enjoyed being petted. I have never worked with a mouse who actively tried to get my attention though. I've always loved mice because they're so darn cute and funny to watch.
- While I haven't had a mouse enjoy petting, I had one who seemed to like eye cleaning as a treatment for sore eyes. In the beginning, I had to scruff her to clean the eyes with saline and a Q-tip. After several days of treatment, the little guy would just sit on my hand and lean into the Q-tip for the cleaning. It probably felt good, so why fight it?
- A long time ago, I had a mother mouse die, leaving behind three 15-days-old pups. I did my best to save them and succeeded. Unfortunately, after all they had been through, they were not suitable to be used for research. Well, I didn't want to euthanize them, so I kept them as mascots. There were two females and one male. I felt bad that the male had no buddy to



live with, so I took it upon myself to be his surrogate. I assume that he came to like my petting, as he would not run away when I put my hand in the cage to rub his head. He would sit in my hand, while I gave him his daily rub, and close his eyes slightly.

- I used to believe that mice do not like to be held, touched or handled. However, during the past year, I have found that it may all depend on who is working with the mice and how they are being handled. Granted, I would say that 90 percent of the mice at my institution would rather be left in their cages 24/7 without disturbance by human hands, but we have a lab here that has truly happy mice who literally buzz when you hold them.

I wouldn't have believed that mice could be that way until I witnessed it with my own eyes. I was so amazed the first time I saw this that the person in charge of the mice placed one of them in my hands so I could feel the buzzing. The little mouse then closed her eyes as I gently rubbed behind her head. She even leaned into the direction of the rubbing so I could get behind her ears. I have since referred to this individual as the *mouse whisperer*; he has taught me that there is much more to the mouse than meets the eye.

- Doesn't your own experience strongly suggest that the way animals respond to us depends primarily on us? After all, mice *are*



social animals, so there is no good reason why they should shun human contact if they can trust the handler.

Some years ago we studied the behavior of a large buffalo herd. Initially, many people warned us not to approach the animals without a fence in between because buffalos are supposedly extremely dangerous and have even killed people. After several days of thorough observations from behind the fence, we climbed over and did our studies while moving along with the animals. Yes, we did follow certain social rules that we had learned during our pilot observations, but were able to take our ethological records directly in the herd. It never happened throughout our two-year study that we were charged or put into any kind of dangerous situation by the animals. No, we didn't try to physically contact them, for the simple reason that buffalo, unlike cattle, do not groom each other, hence probably don't like to be touched.

## *shelter and nesting material*

*There are various plastic shelters—houses, tubes, and igloos—for mice on the market. Based on your own experience, which is the most species-appropriate and practicable shelter?*

- Mice are highly motivated to chew up paper or cardboard to build nests. If this material is provided, I think the pre-fabricated houses are largely redundant. It is the performance of the behavior—building a nest—that is important to the mice, not just its functional consequence of having access to a shelter.
- I would recommend a shelter structure on which the mice can also climb and that has several entrances/exits; the plastic material should be transparent but red-colored so as to avoid disturbing the occupants during the daily checks. Mice appear to prefer shelters without floors. Our mice get shredded paper that they move into the shelter and chew up to form a nest.

We have found that, if you are using plastic shelters, some strains need to quasi-grow up with them in order to use them when they are adults. When we first investigated plastic shelters, we noticed that adult mice rarely used them, but if introduced at the age of weaning, the young mice will accept them more readily and you will end up with almost all adult mice making use of the plastic shelters.

From the mouse's point of view, compressed-paper shelters would be more attractive, but these make the checking of the animals so difficult that we have phased them out in preference for transparent plastic shelters.



- We have never had a problem with plastic tubes for either rats or mice. Yes, the animals can and do chew at the edges of the tube and, after just a week or so, it may need replacing; we have not encountered any teeth or gastrointestinal issues related to that chewing.



- You make a good point about the chewing of plastic; we do see it but, like you, have never encountered any problems as a result of ingestion of plastic particles.

We also use compressed-paper tubes and shelters; this material is always autoclaved first, so it's very unlikely to be the source of any pathogens for the animals.

- We recently began moving away from plastic igloos to commercial paper huts with several openings. Our staff report that the mice seem to prefer the huts. Different strains appear to customize their hut, adding a *sunroof* or a *canopy*, shredding

it completely or using the hut simply as a shelter without damaging it in any manner.

- Taking the standpoint of the mouse, I would certainly prefer reasonably suitable nesting material that I can use to construct a cozy nest at just the right spot of the cage over a prefabricated, heavy, fancy but empty, cold house/igloo that cannot possibly take my personal microclimatic needs into account.
- Van Loo et al. (2005) gave mice a choice of a commercial paper-based shelter (Shepherd Shack™) and a plastic shelter (Techniplast Mouse House™) and noticed that all three strains of mice tested showed a significant preference for the paper shelter. The paper shelter was much lighter (20 g) than the plastic shelter (95 g). This allowed the mice to move it around, manipulate it and change the position of the entrance within the cage. The plastic shelter was probably too heavy for such maneuvering and, hence, never changed its place. The mice also gnawed the paper shelter, occasionally nibbled an extra hole in the side, or shredded part of the walls, using the shreds to strengthen their nest. They could not do this with the plastic shelter. All mice slept inside the paper shelter but never in the plastic shelter. When they slept in the cage that contained the plastic shelter, they did so in the sawdust *outside* the shelter. When tissue paper was provided, the mice dragged the material into the paper shelter and built a

nest, but they never combined this nesting material with the plastic shelter.

- I am not sure there actually is a best plastic shelter; we use several different types that the mice use unpredictably either as a nesting place, a hiding place, or a toilet. What is most important for them, and what they use in a consistent manner is nesting material. Mice love to build nests. When we give our mice Nestlets™ and hay, they typically build a nice igloo-type shelter out of it even if they are also provided with a plastic shelter; I usually find them sleeping in *their* nest but not in the prefabricated plastic shelter.
- I believe that nesting material is more important for mice than a prefabricated plastic shelter. If they have access to nesting material, healthy mice always build a nest and sleep in it. I believe a plastic shelter is useful when several mice are housed together, especially males. If an argument breaks out, the subordinate mouse has a place to hide from the aggressive individual—hopefully out of sight, out of mind. Nesting material doesn't give this kind of cover or protection. We offer both, a cotton fiber pad and a shelter. We use mostly the plastic square houses with several openings. Some mice nest inside the shelter, but many use it to hide from us and from each other, and to climb on. I think both, the cotton fiber pad and the plastic shelter, serve important functions for the mice, so both should be offered to them.

*Is it really important for a mouse to build a nest that serves her as a shelter? Is it not enough to give her a substrate, such as half a cotton fiber pad, that allows her to engage in nest-building behavior?*

- Suppose we place a mouse in an open field, with access to plenty of soft nesting material and an empty plastic shelter, and expose her to a life-threatening stimulus, for example a cat. I would predict that the mouse will ignore the nice nesting material, because the drive to build a nest is temporarily superseded by the survival instinct, but will swiftly escape into the shelter. I would not conclude from the mouse's response that using the shelter is driven by any higher motivation than building a nest, but that the motivation itself is dependent on the context in which a behavior is *needed*.

Mice are pretty much always on the alert for good biological reasons, therefore, I do feel that a mouse should be provided with the necessary substrate that allows her to build a proper—that means closed—nest that can serve her as a safe refuge in the event of danger.

- I think your scenario depends a great deal on the type of nesting material available. If you provide a large amount of shredded paper and a plastic shelter, I predict that,

in response to the cat, the mouse will run under the paper—which unlike the shelter offers many escape points—and take refuge there, without necessarily building a nest at that time.

If, however, you provide a cotton fiber pad and a plastic shelter, I agree with you that the mouse will almost certainly escape into the shelter.

There are many different types of material mice can and will use for nests, from cardboard to electrical cable. Of course, the motivational state for immediate survival (escape from the cat) is different to that of a mouse placed in a cage with little else to do other than perform basic behaviors, such as constructing a nest and improving it whenever new material becomes accessible and can be incorporated.

- In a dangerous situation, mice will go for the shelter, but if the coast is clear they will look for the soft bedding. Case in point is the mice who invaded my garage! I can

see where they live: in the small hole near my septic tank entrance. The mice gained access to my garage and were stealing soft materials and bringing them back to the hole in the ground where they live. I can see pieces of the bedding they bring back; they leave trails of the bedding between my garage and their hole. When they are in the garage, my cat is obsessively staring at the walls. I can watch the mice from my window when they scurry about bringing stuff down the hole. But if I am working around the hole, I never see any of the mice come out; their refuge shelter is then much more important for them than the soft bedding.



*Mice are biologically programmed to build quite elaborate nests that take various behavioral and microclimatic conditions into account. What kind of nesting material is most suitable for mice to build nests in lab cages?*



- Based on my experience with mice, soft paper—not shredded paper—is the most appropriate nesting material for them when kept in cages.

- Van de Weerd et al. (1997) conclude from systematic choice test studies that mice prefer paper-derived materials, such as paper tissues and paper towels, to wood-derived materials such as wood-wool and wood shavings, probably because paper products have a structure that can be converted more easily and appropriately into nests.



- I like shavings along with a cotton fiber pad. It's my experience that mice prefer to build a nest that totally covers them and, when given both materials, most will build this type of nest.

- Nestlets™ are good, but the animals can build much better nests with shredded paper.

- Nestlets™ are 4.8 x 4.8 x 0.5 cm large compressed cotton fiber pads; they are commonly used to offer caged mice the opportunity to build a nest. The nests constructed from one or, even worse, a half of these Nestlets™ have never impressed me, so I am questioning if they can really satisfy the biologically inherent need of a mouse to build a nest.
- In order to cut costs and make mice more visible in research facilities, mice are not given enough material to create a proper nest. A single cotton square is not enough material for a mouse to build a *good* nest, but that is what most mice have to work with.
- Nothing seems to make a mouse happier than building a good nest! But, yes, I agree, the amount of nesting material offered to caged mice is usually pretty scarce.

However, the cage design makes it often problematic to give more. Some mice will construct big nests right next to or around the water sipper tube, a situation that can easily cause flooding of the cage, putting the occupants at high risk if the problem is not noticed and fixed in time. It would be nice to have a cage design for a standard ventilated rack cage that would make it impossible for mice to construct their nests around the water sipper.

- We do use half Nestlets™. We have tried a whole one but the mice love to build their nests under the sipper tube and, by doing so, flood their cage. They never seem happy when wet, so I am thinking they would prefer half Nestlets™ and a dry cage to the wet! Unfortunately, half Nestlets™ are most likely not enough to build a nest in which to hide.

- It seems to me that mice have an extremely strong drive to build a proper, which means closed nest. Whenever a homeless mouse finds suitable or not so suitable material, such as electric wires and some strings under a car's hood, she will not hesitate to build her nest.

It is very strange that nobody has so far come up with a mouse-cage design with a sipper tube that allows mice to build mice-appropriate nests without risk of flooding. Nest building is such an important behavior for mice that, in my opinion, efforts are warranted to more seriously address this behavior in the research lab setting. As a single-caged monkey is not truly a monkey, a mouse without her nest is not truly a mouse.

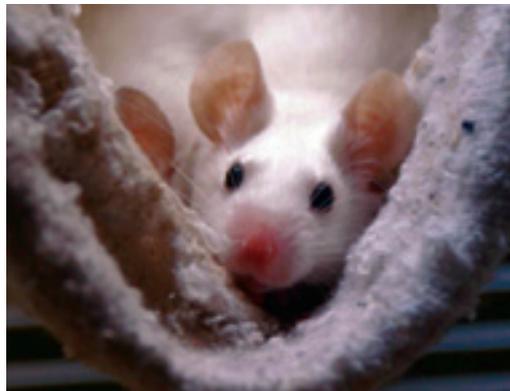


- I have the opportunity to give mice just about every kind of nesting material out there. A combination of soft nesting materials and bedding substrates work best for my mice. They seem to prefer paper towels, but they also will use pre-shredded

paper, crinkle paper (a commercial product), straw and/or cotton together with the paper towel to form a closed nest.

- Many debilitated mice—transgenic, post-surgical, etc.—cannot use some of the commercial nesting products—such as compressed cotton squares—because they do not have the energy to shred this material and build a nest. I have actually seen such mice sitting on top of unshredded Nestlets™! This doesn't provide for thermoregulation, protection from light or other stressors. I think loose cotton or pre-shredded paper should be made available to such animals so that little effort is required of them to build their nests.

- I have also seen some mice who don't know how to pull the cotton fibers apart and end up just sitting on top of the full sized squares. Sometimes, I'll rip the cotton fiber pad and fluff the edges slightly to see if *starting* it for them helps, but then I find them just sitting amongst the pieces I tore. I haven't noted that this lack of proper response is strain-dependent, but have seen mice of different strains sitting on their Nestlets™ rather than using them as nesting material.



*Could it not be that mice need appropriate nesting material, not only to satisfy their strong nest-building drive, but also to construct nests that allow them to conserve heat as needed?*

- Gaskill et al. (2009) report “in laboratories, mice are housed at 20 to 24°C, which is below their thermoneutral zone (26 to 34°C)” and thus, “mice are chronically cold stressed.” If a mouse is permanently kept in a cage that provides a temperature below the animal’s biological thermo-comfort zone, a commercial, small cotton fiber pad—let alone a half one—will not be enough to enable the mouse to build a cozy nest. She will feel always a little bit cold; this, certainly, would not be conducive to the general well-being of the mouse.

*Is wood-wool an enrichment substrate that you would recommend for mice?*

- Mice prefer material that is softer than wood-wool. I have witnessed that mice get their paws entangled in this substrate. It can have sharp parts, causing damage to eyes and paws. The fact that wood-wool is not absorbing is a hygienic disadvantage.
- I agree, wood-wool is not suitable for mice; it not only can cause micro injuries but it is often also very dusty, which may create a health hazard for these small animals.



# blood collection

*From the standpoint of the mouse, what is the least distressing blood collection technique?*

- I have found that mice are less stressed when I take blood samples from the submandibular vein versus the saphenous vein. The mouse is subjected to less restraint, for a shorter time. The vein requires hardly any pressure afterwards to stop the bleeding; this again means relatively shorter restraint time. The vessel bleeds far better, especially in small mice, and a bigger sample can be collected if needed. I prefer to use a relatively large 21-gauge needle, because I think it is less stressful for the mouse to be punctured once with a larger needle than poked several times with a smaller needle to obtain a sufficient amount of blood.
- I agree, the advantages of the mandibular method include:
  - (a) no need for a restraint device,
  - (b) no need to warm the mouse to induce vasodilatation, and
  - (c) the technique is relatively simple and easy to learn, and practical especially when you have to collect numerous blood samples.
- We use the saphenous vein approach with large mice who have a good sized vessel, but I also prefer the mandibular approach with small mice who have tiny saphenous veins.
- With the saphenous collection technique you have to use a lot of caution not to hold the mouse too tightly; over-restraint can cause undue stress or even death. I have seen mice being held so tightly that it injured them while saphenous blood was drawn.
- While there are risks and probably pain involved with any of these techniques, I feel that I have more control and get better results with the submandibular blood collection technique.

I have recently found out how much less stressful it makes the process for the mouse and you if you use the proper lancet. A too large lancet causes too much damage, and with a too small lancet you don't get enough blood. With the correct lancet you can make a good stick one time and get the blood you need quickly, reducing the amount of stress on the mouse *and* you.

- I also prefer the submandibular technique. It is fast, reliable and doesn't require holding off the vessel as does the saphenous technique. Restraint is one of the most stressful things for mice in my opinion. I assume this because they are usually vocal and almost always try to bite!

The saphenous approach is okay, but sometimes the vessel must be held off for a relatively long time and the mice don't seem to like it all that much.

- I would recommend the tail-incision method for blood sampling of conscious mice. The mouse is placed on the cage lid, head and body covered with a tissue. The tail is bent upwards and with a sharp razor blade a perpendicular, small incision is made at the ventral side of the tail about 1.5 cm away from the tail base. Blood drops are then collected in a capillary tube. It is my experience that the mouse is very calm and hardly responds. When finished, a gauze pad is gently pressed on the incision for a moment before the mouse is returned to the home cage.
- Having applied the tail-incision blood collection many times, I can testify that there is no indication that this technique

leads to bone or cartilage damage. However, the incision is so small and superficial that it yields only very small amounts of blood. If larger volumes are needed, I puncture the saphenous vein; for this procedure, the mouse's leg is carefully shaved one day prior to puncture to minimize the stress on the day of the actual blood draw.

- I am going out on a limb and state that I prefer the retro-orbital blood collection technique of conscious mice for the following reasons:
  - (a) done by a trained individual, this technique takes less than five seconds to complete;
  - (b) you can use a microhemocrit tube;
  - (c) if you need to take several samples at different times, the movement of the microhemocrit tube into the canthus is enough to break the clot and allow blood to flow;
  - (d) done correctly this technique does not damage the eye.

Performed by well-trained hands the submandibular, saphenous and retro-orbital blood collection techniques are equal in my opinion. Therefore, if you are the best at retro-orbital, you should use this technique; I certainly do.

- As you say, you are going out on a limb!

How do you measure lack of pain in the mouse when you have to scruff her, holding her so tight that she cannot move? I think the retro-orbital technique should only be done with an anesthetic to alleviate the animal's distress and pain associated with this procedure. This is not a technique for the average vet tech and investigator; it requires extremely skilled and sensitive hands to protect the mouse from serious injury, pain and distress.

- The eye is a particularly vulnerable organ, so just imagining being subjected to a retro-orbital blood collection gives us goose bumps. This is probably the reason why we kind of instinctively infer that a mouse subjected to retro-orbital blood collection must suffer a great deal. It is my experience that this can be the case when the technician or investigator is poorly trained or, even worse, untrained, uses the wrong-size tube, tries to perforate the conjunctiva and is neither careful nor skillful.

I have taken many retro-orbital blood samples from awake mice and never encountered a serious problem. When you know what you do, and how to do it correctly, you can swiftly collect a sample from a conscious mouse without causing damage to the animal's eye and without inflicting undue restraint stress.

- Personally, I see the risk and trauma to the animal during retro-orbital bleeds without anesthesia as unacceptable. I perform several retro-orbitals monthly yet, I still prefer the submandibular. Done correctly on an adult mouse, it is faster and less traumatic than a saphenous, and less risky than a retro-orbital.
- Even when correctly and carefully performed, the retro-orbital approach certainly causes considerable discomfort to the mouse. Although some people use topical anesthesia, I am not so sure this desensitizes all parts involved in this rather invasive procedure.
- Topical anesthesia for retro-orbital blood withdrawal is not enough. That would be only sufficient for anesthetizing the cornea. The pain comes from penetration of the conjunctiva and the deeper tissue layers. To anesthetize these you need to set a retrobulbar anesthesia, which is a rather painful procedure for humans, and probably also for mice. The best option, in my opinion, is a short isoflurane anesthesia for retro-orbital bleeding.

- At the department where I am now working, retro-orbital puncture is routinely performed under light anesthesia. The technicians are very skilled and, therefore, the procedure causes no problems such as hematoma formation after blood collection.

- I've seen seasoned techs who are so fast and precise with the retro-orbital bleed (the mouse is under anesthesia) that for those mice it seemed to be a fairly benign procedure.

I'm not as well-practiced and feel that I cause much more morbidity. I personally prefer the saphenous bleed, especially if only a small volume of blood is needed. The mandibular works well—if you can prick once successfully—but I've seen several mice develop large hematomas afterwards, so I think one needs to hold off for some time after the collection, which might be difficult in an awake mouse.

- Some techniques have an inherently higher risk of doing serious damage, because they involve puncturing vessels in the vicinity of sensitive structures. I am of course thinking of the retro-orbital and submandibular techniques. The great advantages of the saphenous technique are that:

(a) you see what you are doing and the vessel for which you are aiming;

(b) you see what happens after the bleed, for example, if there is any hemorrhage, and

(c) the structures around the vein (skin and muscle) are not so critical.

- Based on my own experience as a veterinarian and based on what I have seen in research laboratories, I would argue that the degree of discomfort (physical restraint) and the intensity of pain (tissue poking, hematoma) and stress (duration of handling) experienced by an animal during blood collection is determined not so much by the technique applied but by the sensitivity and expertise of the person who performs the procedure. If you know how to do a procedure correctly, you probably inflict less discomfort, pain and stress on a mouse during retro-orbital blood collection than when you restrain a mouse incorrectly and have to poke the saphenous vein several times before you can collect a blood sample.

Could it be perhaps that there are not so many people around who do the retro-orbital bleeding correctly as there are people who do the saphenous or submandibular technique correctly?

# aggression

*Male mice can cause aggression-related problems when you transfer them into a new/cleaned cage. What works best, in your own experience, to minimize or avoid altogether inter-male aggression when you clean/change cages?*

- A complete change of the cage bottom reduces aggression; if you then transfer the animals' shelter—or what's left of it—and/or some of their nesting material, fighting will be reduced still further. It seems that some scent markers of the old cage have an aggression buffering effect in the new cage. We transfer our mice directly from dirty to clean cage and see very little fighting; apparently something is working.

If you put the mice from different cages in succession into the same holding container while you are cleaning their cages and then transfer them into clean cages, every mouse will pick up the scent from other cages and you end up with one humdinger of a scrap. I am not surprised that people who do it this way report terrible fighting problems.

I should perhaps add the obvious: we make sure to handle the males before the females or at least change gloves after having handled females.

- We have conducted several experiments and found that the following provisions are

effectively reducing aggression among male mice [Van Loo et al., 2000; Van Loo et al., 2001; Van Loo et al., 2003]:

- (a) keep the number of mice at three per cage;
- (b) provide two shelters per cage (out of sight means, in general, end of fight);
- (c) transfer some nesting material (tissues) from the soiled cage to the clean cage;
- (d) do not transfer bedding material soiled with urine to the clean cage; this would increase aggression.

- It is my practice to transfer the mice along with their old shelter—if it is still in reasonable condition—directly into a clean cage with fresh bedding. The old shelter seems to mitigate overt aggression in the new living quarters.

Our mice always get new Nestlets™ in the new cage. A nest building committee is quickly formed and aggression seems decreased because of the chore at hand.

If the animals get into continuous fighting in the new cage, we don't wait long but check for the primary instigator and remove this individual from the group.

*Could you please share more about your experience of removing the aggressor in dysfunctional groups of mice.*

- It is not always easy to determine the aggressor right away; I must sit and wait, sometimes for quite a while. If I notice squabbling in a cage, I mark it and will then monitor the mice repeatedly on normal days when their cage is not changed. I mark the cage with a sticker, so the researcher or their techs will also be able to monitor the situation and let me know if they catch the problem mouse. The aggressor typically is the guy who is chasing and initiating an argument with the others. Patience may be required to identify him, but it will pay off. If the fighting has gone on for some time, then the guy with the least or no wounds is usually the culprit. Once we have identified the troublemaker, we take him out of the group.

When the rascal is removed, I like to keep the sticker on the back of the cage card so I can easily keep track of that group. Typically, another male will take

the dominant position of the removed aggressor; I have never noticed that the new alpha male acted as aggressively as the old one, so I never had to remove one of them. It seems that the new dominant mouse is just less aggressive, so fights happen less often and they are usually no longer severe and a cause of concern.

- The following quote from a published article (Emond et al., 2003) supports your observation that removing a particularly aggressive mouse can be very helpful to control aggression:

“At our Center, two observation periods were set aside daily in order to identify, according to previously described behaviors, dominant mice and separate these when indicated. By reducing or eliminating the number of aggressive acts between group members in the same cage, our social conflict reduction program has led to a 57 percent reduction of mice being reported for clinical signs, death, and euthanasia.”

- When I wrote an article on this subject some time ago, we were using conventional cages—no filter tops. This made it quite easy to visually and audibly identify dominance behavior whenever it occurred at its early phase.

Since then we have been using filter top cages on ventilated racks. Under this new caging condition, we can barely hear the mice fighting because of the filter top barrier and the noise produced by the ventilators. Additionally, when we do spot an aggressive mouse, it takes a few more steps to first remove the cage from the rack and then take off the filter top and the wire-bar lid from the cage in order to access the dominant mouse. By then, most of the time, we have lost visual contact with the perpetrator and it will take many more minutes of patiently waiting to see if the dominant mouse will act up again—or not—or until it is too late and we find injured animals the next working day.

So, identifying aggressive mice within a ventilated rack system isn't as easy as it is in the old open-cage system described in my article. We are now dealing with more casualties than before.

- Permanent separation of troublemakers is a husbandry intervention that can be useful also in nonhuman primates.

We had serious aggression-related problems with one of our cynomolgus breeding groups. Close observation revealed quickly that there was one particular animal who instigated almost all fights. Removing this individual resulted in a drastic reduction of overt aggression and the group became once again relatively harmonious.

- I had to deal with a similar situation in a rhesus-breeding troop.

There were two female allies—Beta and Witch—who tyrannized most of the lower ranking females of the group. It was quite a terrible situation where we were forced to do something.

After much consideration, I decided to carefully remove the two troublemakers, one at a time, while taking systematic ethological records over a long follow-up period. I spent many hours observing in order to always be prepared for an emergency situation. I was lucky: initially there was a rise in non-injurious aggressive interactions related to a reshuffling of the troop's hierarchical structure. The alpha female and the alpha male kept their positions without any challenges. This probably held the group together; aggressive conflicts quickly subsided and there was no longer any injurious aggression going on. The troop remained stable and compatible for many years [Reinhardt et al., 1987].

*Has anyone had issues with high levels of aggression with FVB mice?  
If yes, what has worked or not worked to decrease aggression?*

- I had serious aggression problems with some FVB males some time ago. I gave them two plastic shelters that have an opening on every side and in the roof, autoclaved hay, a half cotton fiber pad and a bit of shavings. This modification of the males' living quarters reduced overt aggression quite a bit.

- We have very little fighting in our FVB colony; over the years no mouse had to be separated due to fighting or bite injuries.

Our mice are housed in pairs—not trios—in an IVC [individually ventilated caging] system. Each cage has a floor area of 530 cm<sup>2</sup> and is furnished with a cardboard shelter that is open in the front; this allows good visibility of the occupants by the attending technicians and avoids problems with one mouse dominating the entrance. Generous amounts of shredded paper for nesting are also provided. If pups are present, we offer the mice a small amount of pellets on the floor. We use an expanded diet that is very hard; it keeps the mice quite busy. All cages are checked daily and changed once a week.

Originally we worked with FVBs obtained from a commercial source in the United Kingdom. These mice were far more aggressive than our current FVBs obtained from a source in Germany, so there may be a sub-strain problem somewhere along the line.

- Would you please elaborate a bit on the expanded diet.
- This is a diet that has steam pumped through the mix while it is being pelleted. The nutrition is the same as standard extruded pellets but the resulting expanded pellets are very hard and crunchy and a little chunkier. They are more palatable to the mice, and they are microbiologically cleaner due to the high processing temperature. The only word of caution I would add is that, if you have a strain that produces very small and weak pups, some supplementation may be necessary for the first week or so post-weaning. We give ours a few cubes that have been smashed up so that they are easier to chew and ingest.

Most of our feed manufacturers in the U.K. offer both extruded and expanded diets.

*Does access to a running wheel  
-without an attached shelter/igloo- affect intermale aggression?*



- I found that certain hiding items, such as PVC tubes, reduce aggression, but I have witnessed that access to a running wheel actually increases aggression among male mice.
- Our mice have running wheels, but we still see them fighting and have to separate aggressors. Interestingly, the males continue running stereotypically in circles despite having a running wheel.

*Animals tend to compete over a biologically relevant, yet limited resource. Do rodents compete over access to a running wheel?*



- I have never noticed any competition, but many times I have seen up to four mice all running on the same wheel together! Sometimes one of them falls off but jumps back on the wheel immediately without being hindered by the others.

It's the funniest thing to watch. All four mice run on the wheel with their little legs just zooming. If one stops running and the others keep going, the stopped one just spins around on the wheel....weeeee! I also never saw competition, but they are so busy running; I'm not sure they'd be able to fight.

- I have made similar observations in hamsters. Often, two or more of these critters are running in the wheel; no aggressive competition over a space on the wheel, just a lot of hopping on and off.

# barbering

*Partner-directed hair-pulling (barbering) is a common behavior shown by mice kept in laboratories; in fact, it can be so common that it is often accepted as normal for this species. If you see signs of barbering (loss of hair/alopecia) in your mice colony, are you concerned about it?*

- I have mainly noticed barbering in C57BL/6 mice. Most of the time there is one mouse who has all of his/her hair while all other group members are partially bald. Often, if you remove the barber, the others will grow back their hair, but it also happens that one mouse will take on the role of the group's new barber.

I have noticed that each barber has his or her own style, grooming away hair just between the ears or just on the right flank. It seems that individual styles are copied by others. For example, if a flank-barber is placed in a group of mice with an ear-barber, you'll probably end up with mice who will now be missing hair both between their ears and on their flanks.

To be barbered by another mouse doesn't seem to bother the mice overtly. However, if one considers the mouse's sensory musunculus [analogous to the human homunculus which reflects the relative space that parts of the body occupy in the somatosensory cortex; Vanderhaeghen et al., 2000] a large

portion of their sensory input comes from the vibrissae. We could infer from this that, if a barbered mouse lost all of her vibrissae, it might be akin to blinding that important sensory organ.

- Barbered mice are certainly poor choices for any behavioral research, as they will no longer be able to use their whiskers as a biologically crucial source of sensory input for orientation, exploration of the environment, distinguishing objects, locating objects and discriminating the texture of objects. I don't know if the behavioral researchers/investigators are aware of these confounding variables, as many of them were probably taught that barbering was a normal behavior so they could, in essence, ignore it as a confounding variable.
- It is no secret that whisker-barbering will confound an open field test and any behavioral test that requires species-typical sensory capability.

- Barbering/hair-pulling is, in my opinion, a very sensitive indicator that something is seriously wrong with the housing condition of mice or any other species who engages in this compulsive stereotypy. We have become so used to it that we often overlook it as a behavioral problem that needs to be acknowledged and addressed.
- If one is not aware of the fact that a certain behavior, such as barbering, is not normal, then one cannot control for it. In the JAX™ [Jackson Laboratory] mouse info, it says that barbering is a normal display of dominance in mice. Well, if I were an investigator who knows little about mice, I would probably say “hey if JAX says it is a normal behavior then, okay, nothing to worry about; it won’t affect my research because it is a normal mouse behavior.”

I have to fight battles daily with investigators who don’t believe their animals need enrichment because (a) “they are just mice,” (b) “barbering is normal according to JAX” and (c) “barbering does not affect my XYZ research.” I would guess most of the enrichment/behavior folks out there in LAREF land have heard these excuses from not so progressive investigators before.

- It surprises me that no earnest published efforts have yet been made to better understand this behavioral problem, develop refinement strategies to prevent it from developing—at least in those strains of mice in which barbering has no genetic component—and perhaps even eliminate it once it has manifested. Do investigators simply not care, because barbering has no explicit economic implications? In sheep, wool-pulling has economic implications, so strategies have been successfully explored and implemented to control this costly behavioral pathology [Reinhardt, 2005].
- There has been research on barbering in mice, but the problem is that it seems to be a multifactorial phenomenon, with both genetic and environmental factors possibly playing a role. The most important difficulty is that you cannot predict which animal is going to show this behavior; it is also not possible to induce it artificially. This means you cannot set up an experimental design with barbering groups and control groups, with and without environmental modifications, as you can never predict which group—perhaps both or neither—will actually display barbering. So it is not that people are not interested, but more a matter of defining your groups in an experiment.

*From what you have observed, do you get the impression that barbering—similar to hair-pulling in monkeys—is a form of aggression?*

- Although it is often said that barbering is performed by the dominant animal, I dare dispute this. I often observed that not only subordinate but also dominant mice actively offer themselves to get barbered. They would lie down and relax while their cagemate is grooming and barbering them. When the barber stops too soon, he is nudged to resume the barbering.

Of course these are only anecdotal observations, but it would not surprise me if being barbered would activate some endogenous opioid system and, hence, can become addictive. If this is true, the behavior itself may actually be stress-reducing and less of a welfare problem than we think.

- I am also not convinced that the mice on the receiving end are in pain or that the act of barbering is motivated by aggression. I have often seen the animals sit quietly while the barber completes his task. Some of them seem to actively invite the barber to pull their hair. Over time, barbered mice develop bald patches, but this does not seem to affect their physiological and behavioral well-being.

We do identify barbering as an unwanted behavior but we don't regard it as an animal welfare issue.

- Regardless of whether having hairs pulled out is painful or not, having naked patches must hamper thermoregulation for a normally furry animal. Mice who live with a heavy barber may have most of their neck and back plucked.

- The mouse who is being barbered is, in my opinion, not suffering all that much directly from this behavior—apart from possibly feeling a little cold. However, I believe the mouse receiving the barbering is probably suffering because she is experiencing the same inadequate housing conditions that make the active mouse engage in the barbering.

- Rather than focusing on the barbered mouse, we should probably be more concerned about the mouse who does the barbering. After all, barbering is not a normal behavior as it is performed in a stereotypical, almost compulsive manner.

- Barbering mice show histological changes in the prefrontal cortex (Sarna et al., 2000) that are seen also in humans suffering from compulsive hair-pulling, a behavior classified as a mental disorder that can cause clinically significant distress (American Psychiatric Association, 1987). The hair-pulling mouse is perhaps also in a state of distress; if that's the case, barbering would certainly be a welfare concern with possible implications for scientific data obtained from affected animals.

# hamsters



## aggression

*Hamsters can create aggression-related problems when they are caged alone (defensive aggression against personnel) and when they are caged in pairs or groups (injurious aggression against each other). What are practical options to minimize aggression in captive hamsters?*

- Hamsters are nocturnal animals who want to sleep during the day, when we are active. This implies that we are always waking them up during their sleep cycles so, very naturally, they are grouchy. I always give them a minute or two to wake up and only then will I try to handle them. This usually helps to avoid an aggressive overture.

