# Lack of Negative Effects on Syrian Hamsters and Mongolian Gerbils Housed in the Same Secondary Enclosure

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In cases where different species might be housed in the same room or secondary enclosure, the *Guide for the Care and Use of Laboratory Animals* recommends that the animals should be behaviorally compatible and have the same health status. Syrian hamsters and Mongolian gerbils, both desert-dwelling rodents, appear to be reasonable candidates for such a combination. This study was undertaken to evaluate whether housing hamsters and gerbils in the same secondary enclosure is an acceptable practice. Weanling and breeding-age hamsters and gerbils were housed in open-topped cages in an isolator for 5 mo; the isolator also contained with nude and haired mice, which acted as sentinels. Cages housing hamsters and gerbils were rotated between species, and dirty bedding was exchanged between species in an effort to transmit microorganisms. In addition, sentinel mice housed in the isolator were supplied with dirty bedding from both hamsters and gerbils. Neither species showed clinical signs of illness, the health status of neither the hamsters nor the gerbils changed significantly, and the sentinel mice acquired only 2 infectious organisms, a *Helicobacter* species and *Staphylococcus aureus*. Both hamsters and gerbils bred successfully when housed together in the same isolator, and no infanticide or mortality was seen. Breeding performance did not differ between isolator breeding and barrier breeding. This study supports the housing of hamsters and gerbils in the same secondary enclosure.

Although the 8th edition of the Guide for the Care and Use of Laboratory Animals generally recommends physical separation of animals by species, it also mentions that housing different species together in the same secondary enclosure (usually defined as a housing room) may be acceptable when the species are similar in pathogen status and are behaviorally compatible.<sup>10</sup> Syrian hamsters (Mesocricetus auratus) and Mongolian gerbils (Meriones unguiculatus), both desert-dwelling rodents, appear to be reasonable candidates for such a combination. Syrian (or golden) hamsters inhabit extensive burrow systems in the arid, rocky plains of Syria.<sup>5</sup> The burrow systems provide protection from predation and climactic extremes as well as serve as storage sites for hoards of food.<sup>5</sup> Syrian hamsters are solitary animals, with males and females coming together only to mate, and whereas they are nocturnal in the laboratory, these hamsters are diurnal in the wild.<sup>5,6</sup> In comparison, Mongolian gerbils inhabit semiarid, sandy-soiled areas on the Mongolian steppes.<sup>8</sup> Like hamsters, gerbils dig extensive burrow systems, where they hoard food, shelter from predators, and avoid climate extremes.<sup>1,2</sup> Gerbils are diurnal or periodically active.<sup>2,17</sup> Unlike hamsters, gerbils live in social groups.<sup>2</sup>

Charles River Laboratories breeds both gerbils and hamsters at its Kingston, NY, facility, as well as its facility near Lyon, France. According to Charles River's sales figures, the use of both hamsters and gerbils in research has declined from historical levels in both the United States and Europe. These species have been housed in separate rooms in the New York facility, but animal care and husbandry tasks could be accomplished more efficiently by combining the animals into a single room. Hamsters and gerbils inhabit similar habitats in the wild, and the

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2 species have compatible environmental parameters in captivity. Because the native habitats of these species are geographically distant from one another, the predation of one species by the other is unlikely, as is one species serving as a source of stress for the other. Although Charles River's gerbils and hamsters have slightly different health profiles, whether agents that colonize one species infect the other and, if agents are transmitted, whether colonization leads to clinical disease are unknown. Housing the animals closely together in an isolator and evaluating their health, reproduction, and behavior enables the evaluation of potential effects on both species. We tested the hypothesis that housing and breeding hamsters and gerbils in the same secondary enclosure has no observable negative effect on either species.

#### **Materials and Methods**

All work was conducted at Charles River's AAALACaccredited Wilmington, MA, facility and was approved by the IACUC. Nude and heterozygote nude mice (Crl:NU-Foxn1<sup>nu</sup>) used as sentinels were obtained from Wilmington, MA, and were free of many common mouse pathogens; additional details are found at http://www.criver.com/files/pdfs/rms/ hmsummary.aspx. Hamsters and gerbils were obtained from the company's Kingston, NY, facility. The colony of origin for the gerbils was positive for Helicobacter bilis and Staphylococcus aureus. The hamster colony was positive for Campylobacter jejuni, Helicobacter sp. (currently uncharacterized), S aureus, Demodex criceti, Giardia muris, Spironucleus muris, and a Trichomonas sp. Complete health profiles for the hamsters and gerbils are found on the company's website (www.criver.com). All health monitoring was conducted at Charles River's Research Animal Diagnostic Services (Wilmington, MA).

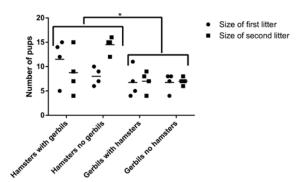
Eight weanling and 20 adult hamsters and 8 weanling and 20 adult gerbils were received from their colonies of origin. The adult hamsters and gerbils were set up as breeding pairs by species. The 8 weanling animals were housed 4 per cage by sex and species.

			Cages fo	or stock produ	ced			Mice
Breeding hamsters	Breeding gerbils	Hamsters	Gerbils	Hamsters	Gerbils			Mice
Breeding gerbils	Breeding hamsters	Breeding gerbils	Breeding hamsters	Breeding gerbils	Breeding hamsters	Breeding gerbils	Breeding hamsters	Mice

**Figure 1.** Physical layout of the isolator, showing the movement of cages and flow of dirty bedding. Double-ended arrows indicate examples of cage exchanges, whereas single-ended arrows indicate examples of dirty-bedding flow to sentinels. Breeding animals are those in pairs already established at the start of the study; no new animals were added to the breeding cohort.

As breeding pairs produced offspring, pups were weaned into cages (maximum, 4 per cage). Breeding pairs remained stable throughout the study. Animal cages were placed in a semirigid, flexible-film-fronted isolator(Ancare RM1, Bellmore, NY), which was 8 feet (approximately 2.4 m) in length. Hamsters, gerbils, and mice were housed in open-topped cages within the isolator. Housing met or exceeded USDA space requirements. The cages contained aspen shavings (Nepco, Warrensburg, NY), and hamsters and gerbils were provided with nesting material (EnviroDri, Shepherd Specialty Papers, Watertown, TN) as well as a structure within the cage (Rat Retreats, Bio-Serv, Frenchtown, NJ). Food (5L79, LabDiet, St. Louis, MO) and water were provided ad libitum; mice were fed from hoppers, but gerbils and hamsters both were fed on the floor of the cage. Water was provided in bottles. The photoperiod was a 12:12-h light:dark cycle, and the temperature was  $21 \pm 1$  °C. In the barrier rooms housing the individual species, a chipped hardwood bedding was used (Beta Chip, Nepco), and no shelters were provided. All other environmental parameters were the same. At every cage change, hamsters and gerbils were provided with cages that had previously housed the other species. Cages were emptied of dirty bedding, but a handful was reserved. The cages were then wiped clean of any residual soil with a dry cloth, and then clean bedding, plus a handful of the dirty bedding from the previous occupants, was added before cages were exchanged between species. Breeder cages were exchanged between breeders, and stock cages were exchanged between stock. Figure 1 provides an overview of the housing schematic and cage exchange. Sentinel mice were given approximately 15 mL (1 tablespoon) of dirty bedding from all cages at each weekly cage change.

The reproductive parameters of time to first litter, interlitter interval, and numbers of pups born and weaned per litter were recorded and compared with those of animals bred similarly in barrier rooms housing these species individually. Hamsters and gerbils on study were observed daily for health and a simple set of behaviors, including stereotypy, infanticide, fighting, and hair loss. The breeding configuration for hamsters and the stocking density for both species differed between the control barrier animals and those on study, so behavior was not compared between the 2. Four animals from each species (2 of each sex from 2 different cages) were tested for the presence of the previously listed microorganisms at 2 time points, that is, 12 and 20 wk after entering the study isolator. At the same time points, 4 sentinel mice—2 homozygous and 2 heterozygous nude mice—were tested also.



**Figure 2.** Analysis of the size of the first and second litters produced by hamsters and gerbils housed with and without the other species. Hamsters have larger litters than do gerbils (P = 0.004). No effect on litter size or any other reproductive parameter measured was seen when animals housed in the same secondary enclosure were compared with those housed in separate secondary enclosures.

All statistical analyses were run by using generalized linear models conducted in Minitab (State College, PA). The assumptions of generalized linear models (normality of error, homogeneity of variance, and linearity) were confirmed posthoc.<sup>7</sup> Significant effects then were analyzed by using posthoc Tukey tests or Bonferronicorrected planned contrasts by using custom contrasts. All values are provided as least-squares means and standard error. A *P* value less than 0.05 was regarded as statistically significant.

## **Results**

Neither stock nor breeding hamsters or gerbils demonstrated hair loss or whisker pulling while housed in the isolator. All adult gerbils in breeding cages performed bouts of stereotypic behavior (corner digging); hamsters showed no stereotypic behavior. No infanticide was seen in breeding pairs of either species. No fighting was seen in single-sex cages of young adult animals of either species. Only one breeding cage of hamsters showed evidence of fighting; in this cage, the male hamster was euthanized for wounds acquired from his mate. These wounds were noted after the birth of the second litter, late in the course of the experiment.

When the breeding performance of hamsters and gerbils housed in the same secondary enclosure was compared with that of the animals housed separately, only litter size differed significantly between hamsters and gerbils (n = 10 cages hamsters, 10 cages gerbils, P = 0.0004, Figure 2). Other measured

 Table 1. Agents present in hamsters before and after housing with gerbils

Agent	Test method	Before housing with gerbils	After housing with gerbils
Sendai virus	Serology	Ν	Ν
Pneumonia virus of mice	Serology	Ν	Ν
Reovirus	Serology	Ν	Ν
Lymphocytic choriomeningitis virus	Serology	Ν	Ν
Simian virus 5	Serology	Ν	Ν
E. cuniculi	Serology	Ν	Ν
B. bronchiseptica	Culture	Ν	Ν
Campylobacter genus		Y (PCR)	N (Culture)
C. coli		N (PCR)	N (Culture)
C. jejuni		Y (PCR)	N (Culture)
C. kutscheri	Culture	Ν	Ν
Helicobacter genus	PCR	Y	Y
H. hepaticus	PCR	Ν	Ν
H. bilis	PCR	Ν	Ν
K. oxytoca	Culture	Ν	Ν
K. pneumoniae	Culture	Ν	Ν
P. multocida	Culture	Ν	Ν
P. pneumotropica	Culture	Ν	Ν
Pasteurella spp.	Culture	Y (NR)	Y
Ps. aeruginosa	Culture	Ν	Ν
Pseudomonas sp.	Culture	Ν	Ν
Salmonella	Culture	Ν	Ν
Staph. aureus	Culture	Y	Ν
Strep. pneumoniae	Culture	Ν	Ν
β <i>Strep.</i> spp.: group B	Culture	Ν	Ν
β <i>Strep.</i> spp.: group G	Culture	Ν	Ν
β <i>Strep.</i> spp.	Culture	Ν	Ν
Lice	Direct exam	Ν	Ν
Mites (Demodex criceti)	Direct exam	Y	Y
Pinworms	Direct exam	Ν	Ν
Protozoa (Giardia, Hexamastix, Spironucleus spp.,	Direct exam	Y	Y
Trichomonads, Entamoeba spp.)			

N, agent was not present; NR, agent was not reported on the health report; Y, agent was present.

Data regarding agents present before housing with gerbils were taken from laboratory results for infectious disease testing for the entire colony. Hamsters, gerbils, and mice typically are tested for different lists of agents, according to customer concerns and species susceptibilities.

parameters (interlitter interval, time to first litter, and interlitter interval) did not differ according to housing condition.

The health status of the hamsters and gerbils did not change significantly over the course of the experiment (Tables 1 and 2). The nude and heterozygote nude mice acquired 2 agents that were not found in their original health profile, a *Helicobacter* sp. and *S. aureus*. (Table 3).

#### Discussion

Housing hamsters and gerbils in the same secondary enclosure in close proximity did not cause distress in either species, according to the behavioral and reproductive parameters measured. Given the natural history of both species and the similar climates in their regions of origin, this outcome is not surprising. Both species share their typical geographic ranges with other rodents, including mice, rats, and other species of cricetid rodents.<sup>15,16</sup> Overlapping ranges of rodent species are common and do not necessarily directly stress animals, as most species have evolved to occupy specific niches (for example, scansorial compared with terrestrial compared with fossorial) or to consume slightly different foodstuffs and thereby mitigate interspecific competition. Although most rodents are somewhat omnivorous,<sup>13</sup> they generally are not considered as strict predators. If they exhibit predatory behavior, it is less likely to be directed at other rodent species but rather at invertebrates and poikilotherms.<sup>3,11</sup> Given the geographic separation of hamsters and gerbils in the wild, it is unlikely that either species would ever encounter the other, so the interaction occasioned by housing in the same secondary enclosure would probably be seen as neutral by both species.

If hamsters and gerbils were housed in the same secondary enclosure, such as an animal production room, implementing spatial and procedural separation would minimize any potential sources of stress. For example, hamsters might be handled by one caretaker on a particular day, with gerbils handled by another, and the species should be housed on opposite sides of the room with a large center aisle between them. Animals undoubtedly would be able to smell the other species and hear their vocalizations, even when the separation within the room did not allow for visual contact. The housing in an isolator is much more intimate and potentially more stressful than in a barrier or experimental housing. In this study, hamsters and Vol 54, No 3 Journal of the American Association for Laboratory Animal Science May 2015

Table 2. Agents	present in	gerbils before	and after ]	housing with	hamsters

Agent	Test method	Before housing with hamsters	After housing with hamsters
Lymphocytic choriomeningitis virus	Serology	Ν	Ν
Clostridium piliformis	Serology	Ν	Ν
B. bronchiseptica	Culture	Ν	Ν
Campylobacter genus		N (PCR)	N (Culture)
C. coli		N (PCR)	N (Culture)
C. jejuni		N (PCR)	N (Culture)
C. kutscheri	Culture	Ν	Ν
Helicobacter genus	PCR	Y	Y
H. hepaticus	PCR	Ν	Ν
H. bilis	PCR	Y	Y
K. oxytoca	Culture	Ν	Ν
K. pneumoniae	Culture	Ν	Ν
P. multocida	Culture	Ν	Ν
P. pneumotropica	Culture	Ν	Ν
Pasteurella spp.	Culture	Ν	Ν
Ps. aeruginosa	Culture	Ν	Ν
Pseudomonas spp.	Culture	Ν	Ν
Salmonella	Culture	Ν	Ν
Staph. aureus	Culture	Y	Y
Strep. pneumoniae	Culture	Ν	Ν
β <i>Strep</i> . spp.: group B	Culture	Ν	Ν
β <i>Strep.</i> spp Group G	Culture	Ν	Ν
β <i>Strep</i> . spp.	Culture	N	Ν
Lice	Direct exam	Ν	Ν
Mites	Direct exam	Ν	Ν
Pinworms	Direct exam	Ν	Ν
Protozoa (Hexamastix)	Direct exam	Ν	Y

N, agent was not present; NR, agent was not reported on the health report; Y, agent was present.

Data regarding agents present before housing with hamsters were taken from laboratory results for infectious disease testing for the entire colony of origin. Hamsters, gerbils, and mice typically are tested for different lists of agents, according to customer concerns and species susceptibilities.

gerbils were housed in cages kept side-by-side, and dirty cages and bedding were exchanged between the 2 species in an effort to transmit microorganisms—2 procedures that would be unlikely to occur in a laboratory or animal production area. We specifically chose the increased stress associated with side-byside housing and cage exchange in an attempt to highlight any negative effects.

The health status of the hamsters and gerbils on study entry was defined according to the results of the most recent overall colony health monitoring for each species. Not every animal in a large colony is positive for all bacterial agents at all times, so the 'loss' of *S. aureus* from hamsters housed with gerbils reflects the status of the founder animals and should not be interpreted as the presence of gerbils eradicating this organism from hamsters. For some agents, testing methods differed depending on laboratory workload and assay development. In the case of *Campylobacter* testing, PCR analysis is more sensitive than is culturing,<sup>12,14</sup> so it is unlikely that animals that were negative by PCR were positive according to culture results.

The only agents transmitted to the nude and heterozygote nude mice were a *Helicobacter* species and *S. aureus*. The *Helicobacter* status of the hamster and gerbil colonies is complicated by the fact that coinfection with various *Helicobacter* species is possible.<sup>4</sup> Hamsters entered the study with at least one unspeciated non-*bilis*, non-*hepaticus Helicobacter*. Hamsters did not acquire *H. bilis* from gerbils. Gerbils already had tested positive for at least *H. bilis* and therefore for the *Helicobacter* genus assay, so it

cannot be ruled out that the unspeciated hamster *Helicobacter* also coinfected the gerbils. Neither species demonstrated any change in clinical condition. In addition, the gerbils acquired *Hexamastix muris*, a protozoan parasite, from the hamsters, but the sentinel mice did not. This outcome may be related to the exchange of both caging and dirty bedding between hamsters and gerbils compared with the addition of dirty bedding only to the mouse sentinel cages. Not every agent monitored would have been transmitted easily to dirty-bedding sentinels,<sup>9</sup> but contact between species that is more intimate than dirty-bedding transfer is unlikely in research and production settings.

Finally, the reproductive performance of the hamsters and gerbils housed in the isolator was compared with that of animals housed in the barrier facility; no reproductive effects were noted throughout the study. The provision of nesting material and a shelter may have served to ameliorate any stress the animals might have experienced and allowed them to cope with their environment. Although all breeding gerbils on study exhibited the common gerbil stereotypy of corner digging, this is not a surprising finding. Corner digging is widely reported among captive pet and laboratory gerbils, can be treated by providing a tube with a 90° bend, and is most likely to be related to the housing provided in the barrier room before shipment.<sup>18</sup> This behavior is seen almost universally in gerbils housed in the barrier room as well, although a prevalence was not recorded for this study, and providing appropriate enrichment is an ongoing project in that room. In summary, housing and breeding

 Table 3. Agents present in mice before and after housing with hamsters and gerbils

Agent	Test method	Before housing with hamsters and gerbils	After housing with hamsters and gerbils
Sendai virus	Serology	Ν	Ν
neumonia virus of mice	Serology	Ν	Ν
/louse hepatitis virus	Serology	Ν	Ν
Ainute virus of mice	Serology	Ν	Ν
/louse parvovirus type 1	Serology	Ν	Ν
/louse parvovirus type 2	Serology	Ν	Ν
linute virus of mice (NS1 protein)	Serology	Ν	Ν
Aouse norovirus	Serology	Ν	Ν
<sup>°</sup> heiler encephalomyelitis virus (GDVII strain)	Serology	Ν	Ν
Reovirus	Serology	Ν	Ν
Epizootic diarrhea of infant mice (Rotavirus: group A)	Serology	Ν	Ν
ymphocytic choriomeningitis virus	Serology	Ν	Ν
Ectromelia virus	Serology	Ν	N
Aouse adenovirus types 1 and 2	Serology	Ν	N
Mouse cytomegalovirus	Serology	Ν	Ν
C virus	Serology	N	N
Polyoma virus	Serology	Ν	N
Jantaan virus	Serology	Ν	Ν
1. pulmonis	Serology and PCR	Ν	N
. cuniculi	Serology	Ν	N
Cilia-associated respiratory bacillus	Serology	Ν	N
Aouse T lymphotropic virus	Serology	N	N
rospect Hill virus	Serology	N	N
bronchiseptica	Culture	N	N
C. coli	Culture	N (NR)	N
C. jejuni	Culture	N (NR)	N
Campylobacter spp.	Culture	N (NR)	N
C. kutscheri	Culture	N	N
<i>lelicobacter</i> genus	PCR	N	Ŷ
I. hepaticus	PCR	N	N
I. bilis	PCR	N	N
. oxytoca	Culture	N	N
	Culture	N	N
P. multocida	Culture	N	N
2. pneumotropica	Culture	N	N
Pasteurella spp.	Culture	Ν	N
Ps. aeruginosa	Culture	Ν	N
Pseudomonas spp.	Culture	Ν	Ν
Salmonella	Culture	N	N
. aureus	Culture	N	Ŷ
trep. pneumoniae	Culture	N	N
Strep. spp.: group B	Culture	N	N
Strep. spp.: group G	Culture	N	N
Strep. spp.	Culture	N	N
.ice	Direct exam	N	N
Aites	Direct exam	N	N
inworms	Direct exam	N	N
Protozoa	Direct exam	N	N
Pneumocystis spp.	PCR	N	N

N, agent was not present; NR, agent was not reported on the health report; Y, agent was present.

Data regarding agents present before housing with gerbils were taken from laboratory results for infectious disease testing for the entire colony of origin. Hamsters, gerbils, and mice typically are tested for different lists of agents, according to customer concerns and species susceptibilities.

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hamsters and gerbils in the same secondary enclosure appears to be an acceptable practice, provided that both species are of similar health status.

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