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HM-28: The Johns Hopkins University Large Animal Enrichment SOP

Performance Goals:

1. To ensure that appropriate enrichment is correctly distributed to all animal species housed at Johns Hopkins University (JHU), excluding mice and rats
2. To ensure that enrichment distribution is correctly documented

Definitions:

ACUC-Animal Care and Use Committee
JHU-Johns Hopkins University
RAR-Research Animal Resources
BMT-Behavioral Management Team

Scope:

This SOP applies to all employees of the JHU RAR and any other personnel working at JHU who distribute enrichment for all animal species, excluding mice and rats.

This SOP is divided into three core sections, supported by information in the appendices:

1. Background information
2. Description of the distribution and logging of enrichment
3. Description of specific enrichment types for large animal species housed at JHU, along with information about the frequency with which each species receives food enrichment

Background

Purpose

This SOP describes the process for distributing non-social enrichment and recording it in the appropriate logs across JHU RAR Central Facilities and best practices for providing social enrichment and to ensure animals housed at all JHU facilities are provided with environments that promote psychological well-being through the expression of species-typical behaviors and that reduce, redirect, and/or eliminate abnormal behaviors.

Animals housed at JHU Central Facilities receive species-appropriate enrichment that meets each species' needs and is in accordance with currently accepted professional standards. Accordingly, this document contains natural history, abnormal behavioral profiles and indicators of compromised welfare, and environmental enrichment information for animal species housed at JHU (Appendix 1). In accordance with Code of Federal Regulations (CRF) 9 Chapter 1 Subchapter A Part 3 Subpart D Section 3.81 (USDA/APHIS), this plan describes how the program addresses the following for nonhuman primates: 1) social grouping, 2) environmental enrichment, 3) special considerations, 4) restraint devices, and 5) exemptions. JHU ensures compliance with all federal, state, and local guidelines and regulations regarding the appropriate care of laboratory animals. Regulations defined in the Animal Welfare Regulations are closely followed to ensure appropriate care for all research animals housed at JHU. JHU is accredited by the Association for the Assessment and Accreditation of Laboratory Animal Care International (AAALAC) and is

therefore in compliance with the *Guide for the Care and Use of Laboratory Animals* (National Research Council, 2011).

Social Enrichment

Interaction with conspecifics and humans are both considered social enrichment. Interaction with conspecifics is regarded as one of the most important forms of environmental enrichment for many species, especially nonhuman primates, though introduction to unfamiliar adults may not be ideal for all individuals or species (NCR, 2011). Unless isolated due to exposure to a contagious disease or for another health concern, social animals must retain visual, olfactory, and auditory access with conspecifics. When appropriate, compatible animals are socially housed with full contact. Individuals may also be socially housed with full contact for breeding and rearing purposes. Human social interaction is attained on a daily basis through husbandry tasks performed by RAR care staff. Additionally, animals may be trained to perform study-related tasks through positive reinforcement.

Non-social Environmental Enrichment

Typically, providing animals with multiple forms of enrichment is the best way to ensure engagement and, in turn, improve welfare. The choice and design of enrichment should be informed by each species' natural history and, where appropriate, tailored to meet the needs of individual animals or study protocols. Additionally, all enrichment provided to JHU research animals must be approved by a veterinarian. There are four broad categories of environmental enrichment, which are not mutually exclusive: food, sensory, cognitive, and physical.

Exemptions

Occasionally, animals may be exempt from any aspect of the environmental enrichment plan due to study requirements listed in the corresponding IACUC-approved research protocol or while under veterinary care due to a medical condition. Nonhuman primates requiring an exemption from the environmental enrichment plan not listed in the IACUC-approved protocol must receive the exemption from the attending veterinarian or their delegate.

While social housing is the default method of housing for many species, some animals may be exempted from social housing (i.e. being co-housed in the same primary enclosure with other animals) due to a number of different reasons as per the JHU ACUC "Social Housing Guidelines for Animals Housed at Johns Hopkins University". All such exemption reasons are marked cage side, in a log, or in the animal's health records.

1. If an animal is not socially housed due to a scientifically justified exemption described in an IACUC-approved protocol, they will be classed as "**Singly Housed: Experimental**".

2. Animals that are singly housed, but *not* due to an IACUC-approved protocol, may be given a social housing exemption by the Attending Veterinarian (AV) or their delegate for one or more of the following reasons:

2a. Veterinary: Animals that are under veterinary care may be exempt from social housing. Depending on their health status, and following approval from the AV, such animals may require to be housed without visual, auditory, or olfactory access to conspecifics.

2b. Quarantine: Animals in quarantine may be singly housed so the animal can be monitored for any disease before (re)joining the population, however social housing should be promoted. Specifically, animals that arrive with known social histories may be socially housed to reduce risk of stress and the aggression that may occur upon reintroduction after an extended period apart. Depending on the health status of such animals, and following approval from the AV, animals may require to be housed without visual, auditory, or olfactory access to conspecifics while being quarantined.

2c. Age: Animals not eligible for social housing due to age.

2d. Behavioral: Animals deemed to be overly aggressive through behavioral observations and/or interactions with conspecifics may be exempt from social housing, as will individuals that show other behaviors that make

them inappropriate for social housing (e.g., they demonstrate inappropriate social interactions with cagemates, or abnormal behaviors that are exacerbated by social housing).

2e. Partner Availability: No compatible social partners are available at the facility. Compatibility may be determined by protocol requirements, breeding requirements, viral status, immune status, or behavioral interactions. Animals that were previously socially housed may also become singly housed due to the death, experimental use, or other status change of their social partner(s).

2f. Timing or Moves: Animals that are about to be transferred to a different facility or enclosure type may be singly housed in that interim period. This includes moves related to internal and external sales.

2g. Breeding: Due to breeding-related requirements animals may need to be singly housed. This includes animals (stud males, pregnant females) that are being rotated between social groups and animals that are required to be singly housed between introductions to new social groups.

3. Animals may be singly housed when they are considered to be eligible for social housing and are currently undergoing social housing introductions. These animals will be classed as “**Singly Housed: Social Status Pending.**” Animals can only be categorized as “Singly Housed: Social Status Pending” while active attempts at socialization are being made. Typically, this would be for a period of no more than three months..

Note about Attrition: As described above, animals may become singly housed due to the death, experimental use, or other status change of their social partner(s). In such cases, the reason of single housing as “attrition” is marked cage side, in a log, or in the animal’s health records. For animals in which socialization with a new partner(s) is likely to be successful, further socialization attempts should be made unless the animal qualifies for one of the above-mentioned exemptions.

Summary of Species’ Enrichment Plans

Table 1.

A summary of the enrichment plan per species.

Species	Food Enrichment		Environmental Enrichment Items		Social enrichment plan
	Distribution frequency	Examples	Number & type required	Examples	
Macaques	3 x per week	Fresh or dried fruits and vegetables, forage mixes, nuts or legumes, manufactured treats	2 medium/large toys per monkey (1 hard, 1 soft)	Hard toy: Large BioServ Dumbbell Soft toy: Medium or Large BioServ Busy Buddy Football	Social housing is default unless there is an exemption or a special consideration. Singly housed animals receive additional enrichment.
Baboons	3 x per week	Fresh or dried fruits and vegetables, forage mixes, nuts or legumes, manufactured treats	2 medium/large toys per monkey (1 hard, 1 soft), plus 1 piece of manzanita wood per cage	Hard toy: BioServ Lucky Clover Soft toy: Kong Dental Dog Toy	Social housing is default unless there is an exemption or a special consideration. Singly housed animals receive additional enrichment.
Marmosets	3 x per week	Fresh or dried fruits and vegetables, forage mixes, nuts or legumes, manufactured treats	Each cage should have 1 hanging hut, 1 piece of manzanita wood, 1 turf foraging board, and 2 small toys (1 hard, 1 soft)	Hard toy: Small BioServ Dumbbell Soft toy: Small BioServ Busy Buddy Football	Social housing is default unless there is an exemption or a special consideration. Singly housed animals receive additional enrichment.
Dogs	2 x per week	Commercially available dog	2 toys per dog (1 hard, 1 soft)	Hard toy: BioServ Nylabone	When cage space allows, social housing is default unless there is a

		treats, dog-safe vegetables (e.g., carrots), peanut butter		Soft toy: BioServ Busy Buddy Football	clinical, behavioral, or scientific exemption. Singly housed dogs have nose-to-nose contact with other dogs through caging when appropriate. BMT staff provide positive human interaction at least twice weekly, including at least one interaction to promote exercise. If at any time a dog is housed in a room without any conspecifics, BMT staff ensure additional daily positive human interaction (as per SOP HM-30).
Cats	2 x per week	Commercially available cat treats	Each cage should have a scratching board, a resting perch, a hanging blanket to hide behind a cat bed, and 2 cat toys The cat room should include additional scratching posts, cat beds, and cat toys	Commercial toys: Small rubber ball, plastic ball with rattle	Social housing is default unless there is a clinical, behavioral, or scientific exemption.
Ferrets	No food enrichment required, but should not exceed 2 x per week if provided	Commercially available ferret treats	Each cage should have a shelter, tunnel, or nest box and at least 2 toys	Small toy: Extra Small or Small Kong, plastic ball	Social housing is default unless there is a clinical, behavioral, or scientific exemption.
Pigs	2 x per week	Fresh or dried fruits and vegetables	3 toys per animal (1 hard floor toy, 1 soft floor toy, 1 hanging toy)	Hanging toy: Porcichew Hard floor toy: BioServ Best Ball Soft floor toy: BioServ DNA Flexer	Social housing is default unless there is a clinical, behavioral, or scientific exemption. Singly housed pigs have nose-to-nose contact with other pigs through caging when appropriate. If at any time a pig is housed in a room without any conspecifics, BMT staff ensure additional positive human interaction.
Sheep and Goats	2 x per week	Fresh or dried fruits and vegetables	2 toys per pen (1 floor toy, 1 hanging toy)	Hanging toy: Porcichew Floor toy: BioServ Best Ball	Social housing is default unless there is a clinical, behavioral, or scientific exemption.
Rabbits	2 x per week	Fresh or dried vegetables, or commercially made treats and forage mixes	3 toys per cage (2 hard floor toys, 1 hanging toy)	Hanging toy: Otto Environmental Stainless Steel Bell Floor toys: BioServ Best Ball, BioServ Dumbbell	Rabbits are typically singly housed but may be socially housed as protocols allow. All animals have olfactory, auditory, and often visual access to conspecifics even when singly housed.
Chinchillas	2 x per week	Fresh or dried vegetables, or commercially made treats and forage mixes	1 shelter per cage and 1 hard toy per animal	Shelter: BioServ Guinea Pig Hut Small hard toy: BioServ Dumbbell	Animals are socially housed when possible. All animals have olfactory, auditory, and often visual access to conspecifics even when singly housed.

Guinea pigs	2 x week	Fresh or dried vegetables, or commercially made treats and forage mixes	1 shelter per cage and 1 hard toy per animal	Shelter: BioServ Guinea Pig Hut Small hard toy: BioServ Dumbbell	Animals are socially housed when possible. All animals have olfactory, auditory, and often visual access to conspecifics even when singly housed.
Woodchucks	No food enrichment required	May be provided with fresh or dried fruits and vegetables, or commercially made treats and forage mixes	1 shelter and 1 toy for gnawing per cage	Shelter: Stainless steel hut provided by breeding colonies Gnawing toy: Manzanita wood	Woodchucks are typically singly housed. They may have olfactory, auditory, or visual access to conspecifics.
Hamsters	1 x week	Fresh or dried vegetables, or commercially made treats and forage mixes	1 shelter	Shelter: BioServ Crawl Ball or appropriately sized mouse or rat tunnel	Hamsters are typically singly housed but may be socially housed as protocols allow. Singly housed hamsters may have visual access to conspecifics.
Bats	No food enrichment required	Not applicable – fresh fruit included in diet as appropriate for species	Each bat room should have roosting shelters, mesh or towels to hang from, artificial tree branches	Commercially available shelters, netting/towels, and artificial plant material	Social housing is default unless there is a clinical, behavioral, or scientific exemption.
Finches	No food enrichment required	Finches may be provided with millet sprays, leafy greens, or other commercially available treats	1 cuttlebone with perch holder per cage, 1 branch/rod perch per cage, 1 flat perch per cage	Commercially available cuttle bones & natural wood perches	Social housing is default unless there is a clinical, behavioral, or scientific exemption.
Owls	No food enrichment required	Dead rats may be provided up to 3 x per week	Multiple perches per cage	Commercially available natural wood perches	Social housing is default unless there is a clinical, behavioral, or scientific exemption.
Turkeys & chickens	No food enrichment required	Turkeys may be provided with leafy greens, grain, or meal worms	Multiple perches per cage	Commercially available natural wood perches	Social housing is default unless there is a clinical, behavioral, or scientific exemption.
Fish, amphibians & reptiles	No food enrichment required	Fish may be provided with brine shrimp	At least one shelter per enclosure	Commercially available shelters	Where possible, fish, amphibians, and reptiles are housed in pairs or groups.

Note. More information about the specific plans for each species are provided in the subsequent sections.

Environmental Enrichment Preparation

- The BMT is responsible for ordering and preparing food enrichment and toys, and for making these enrichment items available to care staff in designated storage areas in proximity to the animal housing areas where they will be distributed.
- The BMT is also responsible for providing reference and documentation materials. The following documents must be kept in, or near, each room housing USDA-regulated animals. These documents may be stored in a green binder labeled “Enrichment Log Book”. If any of the materials are damaged or missing, staff members should report it to a member of the BMT to be replaced as soon as possible.
 - Standard Toy Reference Sheet (for each species housed; Appendix 2)
 - Standard Enrichment Log (for each species housed; Appendix 3)
 - Food Enrichment Guide (for small animal species, e.g., chinchillas, guinea pigs; Appendix 4)
- Care staff supervisors are responsible for alerting the BMT when their areas are running low on toys or dried food enrichment so the BMT may provide additional items. Care staff supervisors are also responsible for alerting the BMT when they begin to house a new species for which they require enrichment materials.

Environmental Enrichment Distribution & Recording Overview

Food enrichment is to be distributed by RAR staff at a frequency determined by the BMT and stated on each species' Standard Enrichment Log. Do not distribute food enrichment to any animals on food restriction. *Note.* Routine food enrichment provision is only required for certain species (as per Table 1) and only those housed at JHU facilities for 15 or more days. All animals are provided with physical enrichment (i.e. toys and other manipulanda).

- The distribution of environmental enrichment must be documented in the Standard Enrichment Log located in or near the animal housing area. The Standard Enrichment Log is typically within an Enrichment Log Book or posted on the wall near the door of each room.
- Standard Enrichment Logs include the following information for reference:
 - The species and the minimum number of times per week that they are provided food enrichment (if at all)
 - The current month and year
 - The corresponding room number or name of the housing area
- If an animal room is empty, the attending staff member should indicate on the log “no animals housed” for the corresponding dates
- At the end of each month, a BMT member reviews the completed enrichment logs, and to do so, may request that staff at each facility send a copy of the logs to them

Enrichment Distribution & Recording Process:

Note. Routine logs of enrichment provision are only maintained for USDA-regulated species housed at JHU facilities for 15 or more days.

The following steps are recommended for distributing and recording enrichment:

1. Confirm that each occupied cage/run has the required number of non-food enrichment toys in adequate condition
 - If a cage/run lacks enrichment or has broken enrichment, replace it. If replacement items are not available in your area, alert your supervisor who will then alert the BMT that additional items are needed.
 - For species that receive food enrichment, this should be done each day food enrichment is distributed. For species that do not receive food enrichment, this should be done once per week.
2. Collect any food enrichment to be distributed that day.
 - When working in an area where food enrichment is prepared daily by the BMT, check the designated area for any food enrichment prepared to go out that day.
 - When working in an area where the BMT has provided a longer-term supply of ready-to-use food enrichment, care staff are responsible for choosing days to distribute those treats to meet weekly minimum frequencies. On days when you are distributing food enrichment, collect treats from the designated storage area.
3. Distribute an appropriate amount of food enrichment per animal.
 - If the BMT has prepared specific amounts for that day, distribute evenly among animals in the housing area, excluding any animals on food restriction
 - If a species has a food enrichment guide provided for them, provide only the amount designated per animal.
4. Fill out the standard enrichment log with the following:
 - A check mark in the column indicating that the standard enrichment items are present and in good condition
 - For species that receive food enrichment, write the name of the type of food enrichment provided in the appropriate column. This can be generalized if care staff are unsure of the exact item (e.g., “greens” can be written for bok choy)
 - The initials of the staff member in the final column for that room
 - Repeat this process for each housing area. One log sheet may include multiple housing areas.

Enrichment for Nonhuman primates

For information regarding the enrichment plan for nonhuman primates housed at the JHU Breeding and Research Farm, see SOP HM-71.

Non-social Environmental Enrichment

Physical Enrichment: In order to provide increased cage complexity, all nonhuman primate cages or enclosures have a variety of elevated perches, hammocks, and /or wooden branches depending on the species. RAR provides primates with two commercially available toys (rubber, hard nylon, etc.) per animal. Nonhuman primates may also be presented with a variety of sensory enrichment that creates visual, auditory, tactile, and/or olfactory stimulation. This may include different substrates to touch and feel (e.g., forage boards), materials with different scents (e.g., frozen ice cubes containing herbs, or cardboard sprayed with an essential oil), or visual stimuli (e.g., videos, bubbles, or moving lights). Such elements will be prepared and distributed by members of the BMT, in consultation with veterinary staff.

Food Enrichment: Each nonhuman primate receives a food enrichment item at least three times per week. These foods are any food other than an animal's standard feed and can include fresh or dried fruits and vegetables, forage mixes, nuts, or manufactured treats. Some nonhuman primates may have exemptions from receiving food enrichment as part of an IACUC-approved protocol. In such cases, behavioral staff work in collaboration with researchers to determine alternative enrichment strategies. For example, nonhuman primates on water regulation programs may still receive food enrichment, but only dried-food options. Alternatively, for animals exempted entirely from receiving food enrichment, behavioral staff provide additional physical and sensory enrichment in addition to the standard toys that each primate receives.

Social Enrichment

All nonhuman primates are housed with visual, auditory, and olfactory contact with conspecifics or members of compatible species, unless there is an approved exemption. To achieve this, the BMT team will identify suitable social partners for each primate. The BMT, in coordination with veterinary and care staff, will introduce individuals to each other through a carefully supervised process. Care is taken to monitor the primates for the expression of positive, affiliative behaviors (lipsmacking, grooming, co-threat, and appropriate dominance interactions such as mounting) and negative behaviors (aggression, biting/fighting, fear grimacing, screaming). After animals are introduced, all staff continue to monitor animals with full contact for the duration of their social housing to assess compatibility. Veterinary staff and members of the BMT should be alerted to any sign of concern, including behavioral indicators (of extreme fear/aggression) and wounding. Typically, pair-housed nonhuman primates are housed in same-sex pairs, but when mixed-sex pairs are formed, a contraceptive plan must also be agreed upon by relevant staff, including veterinary staff, behaviorists, and researchers. In addition to full-contact pair-housing, some primates may have limited tactile contact (grooming holes) with conspecifics in adjacent cages. To promote social housing, exceptions for cage size requirements may be given as described in the JHU ACUC "Social Housing Guidelines for Animals Housed at Johns Hopkins University".

While social housing is the default method of housing for many species, some animals may be exempted from social housing (i.e. being co-housed in the same primary enclosure with other animals) due to a number of different reasons as per the JHU ACUC "Social Housing Guidelines for Animals Housed at Johns Hopkins University".

All singly housed NHP without a social housing exemption approved by the IACUC (i.e. excluding those animals classed as "Experimental") and not in the process of being socially housed (i.e., excluding those animals classed as or "Social Status Pending") will have their exemption status reviewed by the AV every 30 days. In cases where the active socialization process takes more than three months, however, the AV or their delegate must review the status of the animals at that three-month mark to determine if an exemption category is required. To document review, the AV signs the "NHP Social Housing Exemption Log" for all NHPs with an exemption status reason (i.e., "Veterinary," "Quarantine," "Age," "Behavioral," "Partner Availability," "Timing or Moves," or "Breeding") every 30 days.

Special Considerations

As per the Animal Welfare Act, certain nonhuman primates must be provided special attention regarding enhancement of their environment, based on the needs of the individual species and in accordance with the instructions of the attending veterinarian.

1. *Infants and young juveniles*: All infants and young juveniles are socially housed, typically with their dam, unless there is a scientifically justified exemption or veterinary justification. Singly housed infants and young juveniles are closely monitored by personnel each day, provided with soft blankets or veterinary-approved plush toys for comfort, and fed per veterinary instruction. Whenever possible they will be housed such that they can maintain visual and auditory contact with conspecifics.
2. *Those that show signs of being in psychological distress through behavior or appearance*: Such individuals are provided with additional novel foods and/or manipulanda in addition to the standard provision of toys and food enrichment three times per week. The reasons for nonhuman primates to receive additional and/or tailored enrichment include, but are not limited to veterinary requests, behavioral concern, or severe or progressive alopecia (70% or more of the body affected, 50% or more of the body affected with a 25% increase since last score, or majority of areas with alopecia are scored 3/bald). The enrichment plans are developed by the BMT in consultation with the veterinarians and researchers, as needed. The provision and documentation of this additional enrichment provided to nonhuman primates with special considerations is ensured by the BMT.
3. *Those used in research for which the Committee-approved protocol requires restricted activity*: JHU follows the guidelines outlined by USDA/APHIS (§ 3.81) with regard to the use of restraint devices for nonhuman primates, along with the JHU ACUC “Guidelines on Restraining Animals.”
4. *Individually housed nonhuman primates that are unable to see and hear nonhuman primates of their own or compatible species*: All singly housed nonhuman primates, including those that are unable to see and hear conspecifics, are provided with additional novel foods and/or manipulanda in addition to the standard provision of toys and food enrichment three times per week. The enrichment plans are developed by the BMT in consultation with the veterinarians and researchers, as needed. The provision and documentation of this additional enrichment provided to nonhuman primates with special considerations is ensured by the BMT.
5. *Great apes weighing over 110 lbs. (50 kg)*: Not applicable – no great ape species are currently housed at JHU facilities.

Restraint Devices

JHU follows the guidelines outlined by USDA/APHIS (§ 3.81) with regard to the use of restraint devices for nonhuman primates, along with the JHU ACUC “Guidelines on Restraining Animals.”

Behavioral Monitoring

All nonhuman primates are observed by RAR personnel at least once daily including on weekends and holidays. Additionally, nonhuman primates are given a behavioral assessment at least quarterly by members of the BMT. Nonhuman primates exhibiting abnormal/ atypical behaviors (see Appendix 1) are reported to the veterinary staff and behavioral staff, who develop and provide interventions as appropriate. These animals are reassessed during the next behavioral observation. Animals that are observed to exhibit severe abnormal/atypical behaviors (i.e. self-injurious behavior) are reassessed by the behavioral staff on a more frequent basis as determined by the veterinarian and the BMT. All nonhuman primates are assessed for alopecia once per year at minimum by veterinarians and members of the BMT (as per SOP VC-6). The extent of alopecia is calculated using the “rule of nines” approach, and the degree of hair loss is rated on a 4-point scale (Bellanca et al. 2014).

Enrichment for Dogs (*Canis familiaris*)

Social Enrichment

When possible, dogs are pair- or group-housed, with new pair/group formations overseen by behaviorists. All socially housed dogs are closely monitored for any signs of aggression or incompatibility. Dogs may be required to be singly housed due to one of the social housing exemption reasons listed in the JHU ACUC “Social Housing Guidelines for Animals Housed at Johns Hopkins University”. All such exemption reasons are either marked cage side, in a log, or in the animal’s health records. Such singly housed dogs maintain visual, auditory, and olfactory access to conspecifics.

When appropriate, singly housed dogs have nose-to-nose contact with conspecifics through mesh. RAR behavior staff provide positive human interaction at least twice weekly, including at least one interaction to promote exercise. These socialization visits are logged by behavior staff. If at any time a dog is housed in a room without any conspecifics, behavior staff will provide or arrange for additional positive human interaction (as per SOP HM-30).

Non-social Enrichment

All dogs are housed in pens with sufficient space for exercise whenever possible and are provided with at least two commercially available dog toys. Novel treats provided at least twice weekly are distributed in various forms (i.e., hidden in toys, frozen treats, scattered throughout the pen, etc.) by RAR Care Staff.

Enrichment for Cats (*Felis sylvestris catus*)

Social Enrichment

When possible, cats are housed in compatible pairs or small social groups in an activity room that allows opportunities for play and socialization, as well as retreat. Cats living in social pairs or groups are closely monitored for any sign of incompatibility with conspecifics. If needed due to medical or research needs, cats may be housed in cages overnight or for longer periods of time. When housed in cages, cats will have the opportunity to view conspecifics. Care staff interacts with the cats on a daily basis while performing husbandry tasks, and BMT staff provide additional socialization and behavioral monitoring as needed. Cats may need to be singly housed long-term due to one of the social housing exemption reasons listed in the JHU ACUC "Social Housing Guidelines for Animals Housed at Johns Hopkins University". All such exemption reasons are either marked cage side, in a log, or in the animal's health records.

Non-social Enrichment

Cats are housed in, or have access to, cages that have visual blockers for the cats to hide behind. Each cage also has a scratching board, a resting perch, a bed, and at least 2 toys for each cat. The activity room has spaces for cats to hide, numerous scratching boards/posts, cat beds, ledges to rest on, and toys to play with. Cats receive novel treats at least twice weekly. To reduce inter-cat aggression, the cats are provided with individual food and water bowls and multiple litter trays (one in each cat's cage, and additional trays within the activity room).

Enrichment for Ferrets (*Mustela putorius furo*)

Social Enrichment

It is recommended that laboratory housed ferrets are socially housed, especially young, subadult ferrets, females, and castrated males. However, it is important to identify socially compatible social partners, as intraspecific aggression can be a cause of reduced welfare for ferrets. Where possible, ferrets will be pair- or group-housed. In situations where a ferret is housed due to one of the social housing exemption reasons listed in the JHU ACUC "Social Housing Guidelines for Animals Housed at Johns Hopkins University", they will be housed in the same room as conspecifics. All such exemption reasons are either marked cage side, in a log, or in the animal's health records. Ferrets are handled by the care staff prior to and after cage cleaning every other day. During this time, they receive positive human contact through play and petting.

Non-social Enrichment

Ferrets are provided with at least 2 toys to manipulate in their cages. Additionally, ferrets may be offered hammocks, bedding substrate, and/or large huts/shelters/nest boxes to rest in. Ferrets may also be offered access to larger play cages that provide exposure to novel toys, substrates, and space. Ferrets may also be offered commercially available treats up to twice per week.

Enrichment for Pigs (*Sus scrofa*)

Social Enrichment

When possible, pigs are housed in pairs or small groups (although housing them in trios is done with additional care and only in consultation with the BMT). Social groups are typically formed by housing newly arrived animals together (SOP HM-65). When forming new pairs, pairing individuals of relatively similar weights ensures each animal will have access to feed, though dominance rank may take longer to establish. Males over the age of one, who are not already in social pairs, can be more difficult to pair, and tend to show aggressive behavior that can result in

wounding. For this reason, castrated males may be more compatibly housed with female social partners. Sometimes pigs will need to be singly housed due to one of the social housing exemption reasons listed in the JHU ACUC "Social Housing Guidelines for Animals Housed at Johns Hopkins University". All such exemption reasons are either marked cage side, in a log, or in the animal's health records. Singly housed pigs will be housed in a manner that permits animals to have auditory, visual, olfactory, and in most cases snout-to-snout tactile contact with conspecifics. Positive human interaction is received during daily husbandry tasks. If at any time a pig is housed in a room without any conspecifics, behavior staff will provide or arrange for additional positive human interaction, which is logged by the person providing that interaction (care staff or members of the BMT).

Non-social Enrichment

Each pig is provided with one hanging toy and two floor toys, one soft, one hard. They also receive novel treats (fresh or dried fruit or vegetables) at least twice weekly. Treats may be directly handed to the pigs, distributed in their runs, placed in puzzle feeders, or distributed in their feeders so the pigs must use their snouts to root with to free the treats.

Enrichment for Sheep and Goats

Social Enrichment

Where possible, sheep and goats will be pair-housed or housed in small groups depending upon size and social compatibility, and young offspring are housed with the mother. Ideally, sheep and goats should be housed in stable social configurations to reduce stress and aggression. Sometimes sheep and goats will need to be singly housed due to one of the social housing exemption reasons listed in the JHU ACUC "Social Housing Guidelines for Animals Housed at Johns Hopkins University". All such exemption reasons are either marked cage side, in a log, or in the animal's health records. In situations where there is only a single animal of either species, they will be housed in the same room with a compatible species, such as pigs. Additionally, they receive positive human interaction with care staff during daily husbandry tasks.

Non-social Enrichment

In addition to the hay that is provided daily as part of routine husbandry, sheep and goats receive at least two novel food enrichment items (fresh or dried fruit or vegetables) weekly. All pens will include at least one floor toy and one hanging toy.

Enrichment for Rabbits (*Oryctolagus cuniculus*)

Social Enrichment

Rabbits are typically singly housed but may be socially housed as protocols allow. The social housing exemption reason, as per the JHU ACUC "Social Housing Guidelines for Animals Housed at Johns Hopkins University", should be marked cageside. Rabbit cage racks are arranged in a manner that permits animals to have auditory, olfactory, and often visual contact with conspecifics.

Non-social Enrichment

In addition to the hay that is provided daily as part of routine husbandry, each rabbit cage is provided with at least two floor toys and one hanging toy for manipulation. Rabbits receive a novel food item (e.g., fresh or dried fruit or vegetables) twice weekly.

Enrichment for Chinchillas (*Chinchilla lanigera*)

Social Enrichment

Chinchillas may be socially housed as long as they are compatible. Singly-housed chinchillas have auditory, olfactory, and often visual access to conspecifics.

Non-social Enrichment

Chinchilla cages contain a house for them to hide in and sit on and at least one toy per animal. Chinchillas receive novel food items (dried fruit and vegetables) at least two times a week. Daily hay feed may be presented in hanging spiral feeders to encourage foraging behavior. Dust baths are provided weekly as part of routine husbandry.

Enrichment for Guinea Pigs (*Cavia porcellus*)

Social Enrichment

When possible, guinea pigs are socially housed. They are given olfactory, auditory, and often visual access to conspecifics.

Non-social Enrichment

Guinea pigs are provided a CPVC tube or similar structure as a shelter and at least one toy per animal. They receive novel food items (dried fruit/veggies, forage mixes, Timothy hay cubes, etc.) at least twice per week.

Enrichment for Woodchucks (*Marmota monax*)

Social Enrichment

As generally solitary animals, woodchucks are singly housed. When housed in the same room as other woodchucks, they will have auditory and olfactory contact, and they may also have visual contact with conspecifics.

Non-social Enrichment

Woodchucks are provided with a wood shaving bedding to allow for burrowing. They are also provided with a shelter and a toy appropriate for gnawing in each cage, e.g., a piece of manzanita wood or a wooden block. They may be provided with small amounts of food enrichment up to twice per week, such as fresh or dried fruits or vegetables, commercially available treats or forage mixes, or dried browse or flowers.

Enrichment for Hamsters (*Mesocricetus auratus*)

Social Enrichment

As generally solitary animals, hamsters are typically singly housed but may be socially housed as protocols allow. Hamsters are housed singly by default in clear micro-isolators. When housed next to other hamsters, they may have visual contact with conspecifics.

Non-social Enrichment

They are provided with a “Crawl Ball” or tunnel hideout. Hamsters receive novel food items at least once per week, which includes dried fruit or forage mixes.

Enrichment for Bats

Social Enrichment

Bats are socially housed unless single housing is required for scientific, medical, or behavioral reasons. Singly housed bats are housed in rooms allowing olfactory and auditory contact with conspecifics.

Non-social Enrichment

All bats are afforded the opportunity to fly at least three times per week when not hibernating (which occurs October through April). Researchers maintain logs to document that bats housed in smaller cages were provided these flight opportunities. Within their home environment, bats are provided with towels to hide under and bat boxes or crates to roost in. Additionally, branches and netting are provided for the bats to hang from, and from which enrichment can be hung. Some individuals may also be given access to covered, outdoor housing areas.

Enrichment for Birds

Social Enrichment

Where possible, birds are housed in pairs or groups. Socially housed animals are monitored closely for any sign of incompatibility. Mature males might not be pair- or group-housed in order to avoid aggression and injury.

Non-social Enrichment

Multiple perches are made available in all bird enclosures. If nest boxes or shelters are provided to females or breeding groups, multiple should be made available to avoid competition.

Finch cages are equipped with at least one branch/rod-shaped perch and one perch that allows birds to sit with flat feet, e.g., a log slice perch. Perches should be placed at different heights, away from food and water sources, and in a position that allows birds places to sit without their tails touching cage walls. Each cage must also have a cuttle bone hung in a holder with a built-in perch to provide access. Finch cages may also include commercially available hanging toys, lattice balls, or sola balls. Shallow bowls or petri dishes of water may be provided for splashing and bathing.

Finches may be provided with food enrichment up to once per week, including millet sprays, leafy greens, or other commercially available treats. These items are either hung from the ceiling in the corner of the cage, in a feed dish, or on the cage floor to ensure birds have access.

Turkeys and chickens may be provided with food enrichment once per week, comprising of seeds, leafy greens, or mealworms.

Barn owls receive dead rats three times per week, which allows them to exhibit their natural need to hunt and consume prey. Some individuals may also be given access to covered, outdoor housing areas.

Enrichment for Fish, Amphibians, and Reptiles

Social Enrichment

Where possible, fish, amphibians, and reptiles are housed in pairs or groups. Conditions that could require single housing include but are not limited to: incompatibility, disease conditions, and breeding requirements.

Non-social Enrichment

Species that are known to utilize shelter, such as *Xenopus* spp. and catfish, are provided with structures such as artificial plants or plastic tubes. Fish may also be provided with brine shrimp.

Appendix 1: Species' Natural History and Abnormal Behavior

Nonhuman Primates

Natural History

Common marmosets (*Callithrix jacchus*)

Common marmosets tend to live in multi-male/multi-female family groups consisting of 3-15 adults, subadults, juveniles, and infants, in which a single breeding pair is dominant over other group members. Births usually result in twins. Adult offspring of the breeding pair typically stay in their natal group and aid with the care of offspring and territory defense at the expense of their own breeding opportunities (i.e., they experience reproductive suppression). Common marmosets are arboreal and sleep on vines and branches and in tree holes. They are omnivorous, frugivorous, exudativore-insectivores. In addition to using vocalizations to communicate, marmosets display elaborate scent-marking behaviors, used to mark food sources and territory boundaries, and for inter- and intra-group communication.

Reference: Manciooco et al. (2022)

Owl monkeys (*Aotus* spp.)

Owl monkeys are the only nocturnal Anthropoid, but they show peaks in activity at dawn and dusk and show great behavioral plasticity in their activity patterns. They are serially monogamous and live in pairs with up to three offspring. However, in the wild, populations are also comprised of dispersing adults who range solitarily. These single males can be aggressive towards mated pairs, especially when in competition for females. Owl monkeys are arboreal and sleep with their groupmates in sleeping trees during daylight. They are territorial. Owl monkeys use scent cues and vocalization as communication. They are primarily frugivorous, but also feed on saps and insects. For marmosets and owl monkeys alike, given their reliance on scent cues in communication and foraging, when caring for them in captivity it is important to use a sanitization schedule that allows for the transfer of items between cages that carry the "home" scent.

Reference: de la Chica et al. (2022)

Squirrel monkeys (*Saimiri* spp.)

Squirrel monkeys live in large, mixed-sex social groups. They show flexibility and diversity in their social organization: in some species males are dominant over females, in others females are dominant over males, and in others still males and females are co-dominant. Squirrel monkeys show a polygamous mating system and are seasonal breeders. Both males and females disperse from the natal group. Unlike many other primate species, squirrel monkeys rarely engage in allogrooming, and social bonds can be assessed by time spent in social proximity. They use vocalizations, postural displays, and scent marking for communication. Squirrel monkeys are insectivores and frugivores, but have also been observed to opportunistically feed on eggs and lizards, and they spend a very large portion of their day foraging. They are arboreal, highly active, and relatively neophilic.

Reference: Stone and Williams (2022)

Rhesus Macaques (*Macaca mulatta*)

Rhesus macaques live in large multi-male/multi-female groups. Females remain in their natal groups and dominance hierarchy is determined by the dominance level of one's mother. Dominance hierarchy amongst sisters occurs in youngest ascendancy, meaning the youngest outranks her older sisters. Males emigrate from their natal groups prior to puberty. Grooming behavior is used to maintain social bonds and in post-conflict reconciliation. There is a marked breeding season in rhesus macaques, with most births occurring in the springtime. Rhesus macaques communicate through vocalizations and gestures. They are omnivores.

References: Honess (2022), Gottlieb et al. (2017)

Pigtail Macaques (*Macaca nemestrina*)

Pigtail macaques live in large multi-male/multi-female groups. Females remain in their natal groups where dominance is matrilineal, and males emigrate from their natal groups near puberty. Males are dominant over females and often displace them during feeding. Like for rhesus, grooming is an important behavior that secures bonds between individuals. Pigtail macaques communicate through vocalizations and gestures, including an affiliative puckered lip facial expression. Pigtails are not seasonal breeders, but instead mate year-round. They are much less cold-tolerant as

compared to rhesus macaques. The majority of their diet consists of fruits, but they also feed on insects, plants, and small animals.

References: Honess (2022), Gottlieb et al. (2017)

Cynomolgus Macaques (*Macaca fascicularis*)

Cynomolgus macaques live in large multi-male/multi-female groups. Female dominance is matrilineal. Females remain in their natal groups, while males emigrate from their natal groups with peers. Males will attempt to move up in dominance around the age of seven. While cynomolgus macaques have a dominance hierarchy like other macaques, their social system is not as despotic as that of rhesus and pigtail macaques. They communicate through vocalizations and gestures. Cynomolgus macaques are frugivorous, but they will also eat insects, plants, and crustaceans (hence their name “crab-eating macaque”).

References: Honess (2022), Honess (2017)

Bonnet Macaques (*Macaca radiata*)

Bonnet macaques live in multi-male/multi-female groups. Females remain in their natal groups, while males emigrate from their natal groups. Female dominance hierarchy is matrilineal. They are omnivores.

References: Honess (2022), Gottlieb et al. (2017)

Baboons (*Papio spp.*)

Baboons are large and primarily terrestrial, living in a wide range of habitats including semidesert areas, savannah landscapes, rocky hills, and forests and woodlands. Most baboons live in large social groups ranging from 20-200 members. Troops consist of related females, with females inheriting the rank of their mother. Most baboon troops are organized around female-bonded and related matrilineal groups, although the social structure of baboon troops vary by species. Males emigrate from their natal group as they reach puberty. Male dominance is linear within the troop and males are known to be aggressive with each other, but also to form close alliances. Baboons are not seasonal breeders, although some fluctuations in breeding patterns may relate to resource availability. Baboons show social affiliation by time spent in proximity, allogrooming, and lipsmacking. Baboons communicate via complex and varied vocalizations that help coordinate group movement, allow for long-distance contact, and mediate social interactions. Baboons are omnivores, but the majority of their diet is plant material.

Reference: Lutz (2022)

Abnormal Behavior and Indicators of Compromised Welfare

In nonhuman primates, abnormal behavior can be defined as behavior that deviates quantitatively or qualitatively from species-typical behaviors, and they are typically used as indicators of poor welfare (Lutz et al., 2022). Five broad categories of abnormal behavior have been described for nonhuman primates: 1) motor stereotypies (repetitive movements of all or part of the body), 2) self-directed abnormal behaviors (e.g., self-focused behaviors such as eye poking, saluting, self-clasping, and hair plucking), 3) abnormal appetitive behaviors (eating or manipulating feces, urine, hair, or other non-food substances), 4) self-injurious behavior (self-inflicted injuring with the potential to cause harm, or which does result in injury), and 5) withdrawn behavior (a depressive-like state in which the animal shows reduced locomotor activity and typically sits with their head below the shoulders, showing little apparent interest in their environment, but with their eyes open, i.e., *not* sleeping). Abnormal behaviors may reflect a change in welfare, a response to a stressor, or may be a legacy of a previous stressor, impoverished environment or atypical rearing history. Given the range of behavioral expressions and their varied etiologies, behavioral assessments and veterinary evaluations are required to fully interpret the expression of abnormal behavior, and therefore their treatment (Lutz et al., 2022). In addition to abnormal behaviors, nonhuman primates may also express a number of species-specific indicators of arousal, fear, or pain, which should also be monitored. For example, in marmosets, freezing behavior (motionless, with ear tufts in a downward position, and eyes partially closed) is a sign of fear and anxiety. Similarly, marmosets show increased rates of the species-typical scent marking behavior in response to environmental stressors (Manciocco et al., 2022).

Ethogram developed by the NPRC Behavioral Management Consortium. Available at: <https://nprcresearch.org/primate/behavioral-management/abnormal-behavior-ethogram.php> [accessed July 14 2022].

- Bizarre posture:** Holding a seemingly uncomfortable or contorted position
- Bob:** A rapid and repetitive* up and down motion of the body on flexed limbs; animal does not leave the cage surface
- Bounce:** Repetitively* using one's hind legs or all four limbs to push oneself off the cage surface
- Coprophagy:** Ingesting feces or manipulating feces in the mouth
- Eye poke/ Salute (Periorbital contact):** Animal holding hand, digit, and/or object against/near one's eyebrow or eye
- Feces paint:** Smearing and/or rubbing feces on a surface
- Flip:** Repeated forward or backward somersaults, may utilize the cage sides or ceiling
- Floating limb:** An arm or leg rises into the air and may or may not contact the body (e.g., gently stroking the body). The action appears to be non-volitional; the animal may interact with the limb as if it is not part of the body. This behavior may be associated with SIB such as self-biting or self-hitting
- Food smear:** Spreading of chewed food on a surface with the mouth; food is often licked off
- Hair pluck:** Removal of hair from one's own body by pulling with teeth or hands, often seen with a quick jerking motion
- Head banging:** Repetitively* and forcefully hitting the head against an object or surface
- Head toss:** Repetitively* moving head side to side, or in a circular manner
- Pace:** Repetitive* locomotion following the same path - for example, walking back and forth on the ground, around the enclosure, or back and forth across bars
- Regurgitate:** The backward flow of already swallowed food - the material may be retained in the mouth or deposited on a surface and re-ingested
- Repetitive licking:** Prolonged or excessive contact of tongue with a surface or object for no apparent reason
- Rock:** Any rhythmic motions of the body from a stationary position. Animal remains sitting or standing while the upper torso sways back and forth
- Self-bite:** Closing teeth rapidly and with force on oneself
- Self-clasp:** Clutching one's own body with hands or feet
- Self-injure:** Any behavior by the animal that causes physical trauma to itself such as bruising, lesions, lacerations, or punctures
- Self-oral:** Sucking a part of one's own body
- Self-slap:** Forcibly striking oneself with hands or feet
- Spin:** Repetitive* circling of body around a pivot point
- Urophagy:** Licking or ingesting urine
- Withdrawn:** Slumped or hunched body posture, often accompanied by dull eyes, and relatively unresponsive to environmental stimuli to which other monkeys are or typically would be attending
- Other stereotypical locomotion:** Idiosyncratic repetitive* whole body movements, particular to an individual; does not meet criteria for other behaviors defined above
- *Repetitive = a minimum of 2 or 3 times, depending on your facility's criteria

Dogs

Natural History

The domestic dog is the only fully domesticated species from the family Canidae. Dogs are social animals and live in fluid social groups that do not form strict hierarchies. All group members may breed, and females typically come into estrous twice a year. Dogs are opportunistic feeders who rely on scavenging. Dogs communicate via a range of vocalizations, and they also rely on olfactory cues to interpret their social and physical environment. Socialization from an early age is necessary for dogs to develop appropriate behaviors for interacting both with other dogs and people. They benefit from positive human interaction, consisting of petting, praise, and treats, and participating in positive reinforcement training.

References: Hall and Prescott (2022), Serpell (2017)

Abnormal Behavior and Indicators of Compromised Welfare

Poor welfare in dogs is typically expressed via changes in the rates or duration of species-typical behaviors, rather than the presentation of unique "abnormal" behaviors (Hall & Prescott, 2022). For example, while brief bouts of

vigilance behavior (standing alert) may be seen in bouts of play, prolonged vigilance behavior is a welfare concern. Other indicators of high arousal include pacing, jumping, and standing on their hind legs with their front paws against the enclosure wall. Acute stress responses, often in response to a change in social or physical environment, or the presence of an unfamiliar person, include excessive licking, panting, or cowering.

Cats

Natural History

Although cats have been kept as pets for thousands of years, they are still not considered fully domesticated. Cats show great plasticity in their social organization, and they can live well in social situations but are known to easily adapt to solitary living. Both scent markings and visual cues are important forms of communication, as are vocalizations. Visual communication is commonly used to modulate agonistic interactions (arching their back, side-to-side twitching of the tail tip) or indicate fear (low, crouched posture with flattened ears). However, affiliative body postures are also seen (stretching, allogrooming, or headbutting), and such behaviors can also be directed at humans as well as conspecifics.

References: Driscoll et al. (2009), Stella (2022)

Abnormal Behavior and Indicators of Compromised Welfare

Cats often seem to express few or subtle signs of welfare problems. Common responses to chronic stress include decreased appetite, vomiting, urinating and defecating outside of the litter tray, decreased social behavior, decreased self-grooming and care, and increased lethargy and attempts to hide. Cats also show a number of postural changes indicative of stress, fear, or pain (Rochlitz & Yeates, 2019), which can be assessed using the Cat Stress Scoring Scale (Kessler & Turner, 1997). Cats produce a range of vocalizations that can indicate negative affective states (yowls, snarls, hisses, and shrieks). Cats may purr when being groomed, but also when fearful or in severe pain, so purring is not a reliable sign of contentment. Cats in acute pain may vocalize (growling, hissing, spitting, yowling) or be silent. When in pain, they are also often withdrawn, inactive, and tense, with a reduced appetite and changes in demeanor such as aggression or extreme timidity. A crouched sternal posture with a stiff, ventro-flexed neck, or other abnormal postures may also be seen (Rochlitz & Yeates, 2019).

Ferrets

Natural History

Ferrets are fully domesticated. They are strictly carnivores, with the inability to digest plant matter. They are naturally solitary and highly territorial, using the scent glands located on the anus to mark territory. However, when housed with socially compatible partners, they can form strong affiliative social bonds, and they sleep in small spaces huddled together. Indeed, research has shown that ferrets will actively seek out social partners when housed in laboratory settings and that they preferentially choose to spend time near their social partner. Ferrets are very curious and neophilic and will engage in frequent bouts of physical, social play when housed socially, as well as play with toys and objects. They make a loud chuckling sound and their bodies shake when excited. Ferrets seek out enclosed spaces to sleep and hide in, and so providing them with tunnels, hammocks, or substrates in which they can bury themselves can be beneficial.

References: Jimenez et al. (2023), Reijgwart et al. 2016, Vinke et al. (2022)

Abnormal Behavior and Indicators of Compromised Welfare

When housed in a suboptimal environment, ferrets may show changes in the rates of species-typical behaviors, which can indicate compromised welfare (Vinke et al., 2022). For example, they may engage in excessive scent marking or intraspecific aggression. Changes to the physical complexity of their home cage, changes in social partners, or an increase in positive interactions with care staff can help to address such territorial behavior concerns. Additionally, given ferrets' curiosity and intelligence, a lack of mental and physical stimulation can result in locomotor stereotypies, stress-induced alopecia, destructive behaviors, hyperactivity, or increased lethargy. These maladaptive behaviors can develop into chronic abnormal behaviors. Behavioral indicators of pain or distress include anorexia, lethargy, immobility, a hunched posture, whimpering, or an absence of self-grooming. (However, it is important to note that ferrets often walk and play with an arched back, and this should be differentiated from the aforementioned hunched posture.) Teeth grinding may also be observed, and a facial grimace scale for pain has also been developed for ferrets (Vinke et al., 2022).

Swine

Natural History

Pigs are social animals that live in large groups. Among littermates, dominance is established within the first two days following birth, with the largest and firstborn often establishing the dominant rank. Unfamiliar pigs will establish dominance within a few days of introduction. Aggression has been noted during feeding, with dominant animals monopolizing the majority of food. In the wild, pigs live in large, complex environments and spend the majority of the day foraging and exploring, as well as rooting through dirt and other substrates. They rely heavily on olfactory cues, more so than visual cues, and their sensitive snout helps them when foraging.

References: Edwards and Grand (2022), Kittawornrat and Zimmerman (2010)

Abnormal Behavior and Indicators of Compromised Welfare

Tail biting and ear chewing of pen mates, as well as chain chewing and vacuum chewing, can occur in socially housed pigs that lack the ability to root. In barren environments, these behaviors can become stereotypical, repetitive behaviors (Edwards & Grand, 2022). In pigs, the presence and degree of tear staining can also be used to assess welfare. Tear stains can be rated on a 0 (no staining) to 5 (staining is severe and extends below the mouth line) (Telkänranta et al., 2016). Pigs in poor environments show more severe tear staining.

Sheep and Goats

Natural History

Sheep (*Ovis aries*)

Sheep live in social groups known as flocks. They are relatively defenseless against predators – they typically respond to threats by fleeing. Sheep benefit greatly from social support and form strong bonds with flockmates. They can be fearful animals, and so habituation to care staff via positive interactions is essential. The social organization of sheep follows a matrilineal structure, with one or two dominant individuals that dictate group movement and foraging patterns. Sheep are herbivorous ruminants and spend a large period of their time grazing and ruminating.

Reference: Dwyer (2022)

Goats (*Capra hircus*)

The domestic goat is a sociable, inquisitive, and intelligent species, although they can be aggressive both towards conspecifics and human care takers as a means to establish dominance. Goats are social and benefit from living with conspecifics. In goats, more than in sheep, olfactory signals are important, especially in sexual and maternal behavior. Like sheep, goats are herbivorous ruminants and spend a large period of their time grazing and ruminating.

Reference: Miranda-de la Lama and Mattiello (2010)

Abnormal Behavior and Indicators of Compromised Welfare

Abnormal or stereotypical behavior is not commonly observed in sheep and goats, although this may mean that non-behavioral indicators of poor welfare need to be determined (Tamioso et al., 2017). However, singly housed individuals have been reported to perform stereotypical oral behaviors, such as chewing pen fixtures, mandibulation, and repetitive licking. Locomotor stereotypies have also been observed, which include rearing, repetitive headbutting, arching the head and neck over the back (“star-gazing”), as well as pacing and weaving (Dwyer, 2022). In sheep, ear position can be used as indicator of in-the-moment welfare state (Boissy et al., 2011). When both ears are in the plane position (the two ears are in the frontal plane and auricles are concealed from frontal view), a neutral mood is inferred. However, when the ears are in a backward position or in an asymmetric position, the animal is likely experiencing some negative event or stressor.

Rabbits

Natural History

Rabbits are social animals. Adult males tend to be more aggressive than females. Female rabbits are easier to socially house than males. Though nocturnal by nature, rabbits housed in a laboratory setting often following diurnal behavioral patterns. Rabbits have a strong prey instinct and can become easily scared. They are herbivores, consuming a diet higher in protein and soluble carbohydrates. Rabbits produce a night feces rich in nutrients, which is ingested.

References: Suckow et al. (2012), Suckow and Douglas (1997)

Abnormal Behavior and Indicators of Compromised Welfare

Animals in suboptimal conditions that do not allow for the full expression of species-typical behaviors can develop abnormal behaviors, such as excessive wall pawing and bar gnawing (Lidfors & Dahlborn, 2022). Rabbits can also show self-directed abnormal behaviors, including repeated self licking, sham chewing, and self biting. Stereotypic behaviors appear most frequently at night when rabbits are more active, so they can be missed via daytime observations.

Chinchillas

Natural History

Chinchillas are very active animals that tend to be shy and easily frightened. They live in large groups consisting of many smaller family groups. Females are dominant over males and tend to be more aggressive. They are nocturnal by nature but adjust to a diurnal lifestyle in the laboratory. They are considered herbivores but occasionally eat insects. They dust bathe in order to maintain a healthy coat.

Reference: Suckow et al. (2012)

Abnormal Behavior and Indicators of Compromised Welfare

The most common abnormal behaviors observed in small mammals, such as chinchillas, include self-directed behaviors, such as excessive grooming, hair plucking (barbering), fur chewing, and self-biting; stereotypical movements, such as somersaulting and digging; and chewing and mouthing of caging (Vergneau-Grosset & Ruel, 2021). Such behaviors can be seen in response to pain (both acute and chronic), stress (both acute and chronic), and an impoverished environment, which can lead to boredom.

Guinea Pigs

Natural History

Guinea pigs are very docile rodents that prefer physical contact with conspecifics. Guinea pigs do well when socially housed; however, males can become aggressive with other males. They are thigmotactic, but will venture into the center of their cage if burrowing material or shelters are available. Guinea pigs are sensitive to noise and show a strong anti-predator response. Guinea pigs, like other rodents, have teeth that constantly grow and so should be provided with material to chew on, otherwise they may exhibit cage biting.

References: Kleven (2022), Suckow et al. (2012)

Abnormal Behavior and Indicators of Compromised Welfare

Loud, sudden noises can startle guinea pigs. They respond by trying to flee or freezing. Freezing can last from a few seconds to several minutes. Guinea pigs also show tonic immobility in response to restraint. Increases in both freezing behavior and tonic immobility suggest a high-stress environment (Kleven, 2022). Cage biting is another common behavior, especially when chew toys or wooden blocks are not provided. Similarly, repetitive water bottle manipulation may be seen.

Woodchucks

Natural History

Woodchucks are large rodents that raise their young and hibernate through winter in extensive underground burrows. Providing opportunities for digging behavior and a burrow-like structure is therefore critical to behavioral health in captivity. Woodchucks are mostly herbivorous, eating vegetation, flowering plants, berries, and roots in the wild. They live alone, except during breeding and while females raise young.

References: Alaska Department of Fish and Game: Woodchuck species profile (n.d.), Bellezza et al. (2015)

Abnormal Behavior and Indicators of Compromised Welfare

When alarmed or distressed, woodchucks may utter a loud whistle. They may also hiss, squeal, and growl. Woodchucks are quite active animals, but lethargy and anorexia are seen in both sick animals and torporous animals preparing for hibernation. Other clinical and behavioral measures must therefore be used to diagnose illness. Aggressive behavior in males may increase when housed in proximity to females during breeding season.

References: Alaska Department of Fish and Game: Woodchuck species profile (n.d.), Bellezza et al. (2015)

Hamsters

Natural History

In the wild, hamsters are diurnal, but in captive settings, they are nocturnal animals and spend a significant amount of time sleeping. Aside from when foraging for food, hamsters typically spend time in burrows. Hamsters are territorial, and both sexes mark their territory using flank glands and defend their territories aggressively. Indeed, adult hamsters are typically solitary and only come together to mate. Single housing may be required when housing adults because unfamiliar adults can be quite aggressive towards each other; however, litter mates or animals introduced at an early age tend to get along well. Hamsters, both male and female, build nests. They are granivorous, but will also eat plants, insects, and fruits. Given hamsters' need to burrow and hoard their food, providing them with deep-litter bedding is advisable.

References: Suckow et al. (2012), Winnicker and Pritchett-Corning (2022)

Abnormal Behavior and Indicators of Compromised Welfare

The most common abnormal behaviors observed in small mammals, such as hamsters, include: self-directed behaviors, such as excessive grooming, hair plucking (barbering), fur chewing, and self-biting; stereotypical movements, such as somersaulting and digging; and chewing and mouthing of caging (Vergneau-Grosset & Ruel, 2021). Such behaviors can be seen in response to pain (both acute and chronic), stress (both acute and chronic), and an impoverished environment, which can lead to boredom. Handler-directed aggression may also be observed, especially when trying to handle them during the day (when they are sleeping) or if they have not been appropriately socialized. Approaching hamsters slowly and using the scoop technique, rather than scruffing, can reduce aggressiveness when handling (Winnicker & Pritchett-Corning, 2022).

Bats

Natural History

Short-tailed Fruit Bat (*Carollia perspicillata*)

Short-tailed fruit bats have a heightened sense of smell used in locating food. They leave their day roost after dusk to feed on fruits and seeds. During feeding, they take meals to a feeding roost, which is different from their day roost. Short-tailed fruit bats live in social groups, consisting of one male and one or more females with offspring. Males that do not yet have their own breeding group live in all-male bachelor groups. Males are territorial and protect their breeding groups.

References: Altringham (2011), Fascione (1995)

Egyptian Fruit Bats (*Rousettus egyptiacus*)

Egyptian fruit bats use echolocation while flying in order to avoid obstacles in their path. Their diet consists of a variety of fruits, flowers, and leaves, and they rely on their sense of smell in selecting food. They roost in large groups and compete for the darkest location within their roosting spot. While roosting, they maintain close contact with conspecifics. During the night, while they are active, they may spend up to half of the evening grooming food from their hair.

References: Altringham (2011), Fascione (1995)

Big Brown Bats (*Eptesicus fuscus*)

Big brown bats rely on echolocation while flying to avoid obstacles and locate insect prey. Females roost together in the spring/summer (after hibernating) in groups referred to as "maternity colonies." Males tend to roost alone, but they have been known to roost with either sex. Big brown bats tend to hibernate individually or in small groups of just a few individuals.

Reference: Altringham (2011)

Abnormal Behavior and Indicators of Compromised Welfare

Changes in body condition, skin condition, and weight are monitored as indicators for changes in welfare, as are behavioral indicators, such as increases in lethargy (although this is seasonally variable too), decreased interest in training and food, and increases in activity. For breeding females, repeated abortions are also a reliable indicator of poor welfare. A variety of physical and social enrichment opportunities can help promote species-typical behaviors and enhance welfare (LeBlanc, n.d.).

Birds

Natural History

Barn Owls (*Tyto Alba*)

Barn Owls are nocturnal predators that hunt small mammals and occasionally small birds. They use eyesight and hearing to find prey, though, if necessary, they can rely solely on hearing to locate and capture prey. They are known to be monogamous, but can be polygamous.

Reference: All About Birds: Barn Owl (n.d.)

Japanese Quail (*Coturnix japonica*)

Quail are social animals with well-defined hierarchies. They are also territorial and mark their territory by vocalizing from an elevated location within their territory. It can be difficult to introduce unfamiliar animals to established groups, and this usually results in aggression and fighting. Quail are ground-feeding omnivores. They are observed to spend time ground scratching, ground pecking and dustbathing.

Reference: Dixon and Lambton (2022)

American Singer Canary (*Serinus canaria*)

Canaries are social animals living in large groups and benefiting from social housing. Multiple females can live together in an enclosure, though males can be territorial and may require single housing to avoid fighting with other males, depending on cage size. Males attract mates through singing.

Reference: Coutteel (2003)

Zebra Finches (*Taeniopygia guttata*)

Zebra finches are gregarious and can live in flocks numbering over 100 individuals, although they typically spend most of their time in pairs or small groups. Indeed, they form long-lasting pair bonds. Flocks show high synchronization in the performance of daily behaviors, such as feeding, roosting, and preening. Zebra finches are very vocal and their varied calls allow for both intra- and inter-group communication.

Reference: Friedrich and Mello (2022)

Domestic chickens (*Gallus gallus domesticus*)

Fowl are a very social species and live in flocks, which are structured following a dominance hierarchy. They spend a large part of their time foraging for food and, in captivity, they are motivated to perform this behavior even if food is freely available. Chickens perform dustbathing behavior, which helps remove debris and ectoparasites from their feathers. Sexually mature animals also seek out opportunities to nest and the provision of nest boxes and perches is beneficial to their welfare.

Reference; Dixon and Lambton (2022)

Turkeys (*Megeagris spp.*)

Turkeys are group-living birds with a highly competitive social system. Dominance hierarchies are often related to kinship and unrelated mature males are often the recipients of aggression from in-group members. Accordingly, stocking density in captive settings is an important factor for welfare. Feather pecking and aggressive interactions should be monitored closely as potential indicators of social incompatibility.

Reference: Marchewka et al. (2013)

Abnormal Behavior and Indicators of Compromised Welfare

The captive environment may elicit increased levels of aggression among birds, especially males. While minor feather plucking is a normal component of preening, excessive plucking that results in bald patches is indicative of poor welfare (van Hoek & Cate, 1998). Birds may overpluck their own feathers, or those of cagemates. Birds that are typically very active and vocal (e.g., zebra finches) that show increased inactivity or a reduction in song production may also be experiencing poor welfare. Route tracing is also seen in captive canaries (Engebretson, 2006).

Fish, Amphibian, and Reptiles

Natural History

Fish (Various spp.)

The species of fish used in research at Johns Hopkins range from highly social “schooling,” open water species such as *Danio* spp. to less social bottom dwellers such as catfish.

Reference: Fox et al. (2002)

Frogs (*Xenopus* spp.)

Xenopus are completely aquatic and, like most amphibians, seek shelter as a means of predator avoidance.

Reference: Fox et al. (2002)

Anoles (*Anolis* spp.)

Anoles regulate their body temperature by shifting between sun and shade. Male anoles are territorial and will form dominance hierarchies if group housed, requiring housing to provide multiple points of access to light and shade.

Acute stress can cause a dark eyespot to become visible.

References: Poole (1999), Fox et al. (2002)

Appendix 2: Example of a Standard Toy Reference Sheet

Standard Toys for Macaques

Each **Macaque Cage** Should Have...



1 Medium/Large Hard Toy **per monkey**



1 Medium/Large Softer Toy **per monkey**

Appendix 3: Examples of Standard Enrichment Logs

Standard enrichment log for a housing area with multiple rooms that receive food enrichment

Standard Enrichment Log				Swine (2x/week)		Nov. 2023	
Ross 429A				Ross 429B			
Day	Confirmed correct number & type of toys were present and in good condition? If missing or in poor condition, added/replaced appropriate toys? (Check)	Type of food enrichment provided	Initial here	Confirmed correct number & type of toys were present and in good condition? If missing or in poor condition, added/replaced appropriate toys? (Check)	Type of food enrichment provided	Initial here	
1	✓	Pear	SH	✓	Pear	SH	
2							
3							
4							
5							
6	✓	Carrots	JS	✓	Carrots	JS	
7							
8	✓	Zucchini	SH	✓	Zucchini	SH	
9							
10							
11							
12							
13	✓	Brussel Sprouts	JS	✓	Brussel Sprouts	JS	
14							
15	✓	Carrots	JH	✓	Carrots	JH	
16							
17							
18							
19							
20	✓	Cucumbers	TL	✓	Cucumbers	TL	
21	✓	Sweet potato	MP	✓	Sweet potato	MP	
22							
23							
24							
25							
26							
27	✓	Cucumbers	TL	✓	Cucumbers	TL	
28							
29	-	Pineapple	JH	-	Pineapples	JH	
30							






Standard enrichment log for a housing area with a single room that does not receive food enrichment

Standard Enrichment Log		Finches	Sept. - Dec. 2023
Blalock 1323A			
Week of	Confirmed standard enrichment items (1 cuttlebone per cage) were present and in good condition? If missing or in poor condition, added/replaced appropriate items? (Check)	Initial here	
Sept. 4	✓	MB	
Sept. 11	✓	MB	
Sept. 18	✓	MB	
Sept. 25	✓	DB	
Oct. 2	✓	DB	
Oct. 9	✓	DB	
Oct. 16	✓	DB	
Oct. 23	✓	DB	
Oct. 30	✓	DB	
Nov. 6	✓	DB	
Nov. 13	✓	DB	
Nov. 20	✓	DB	
Nov. 27	✓	DB	
Dec. 4	✓	DB	
Dec. 11	✓	DB	
Dec. 18	✓	DB	
Dec. 25	✓	DB	

Appendix 4: Example of a Food Enrichment Guide

Guinea Pig Food Enrichment

Food enrichment should be provided at least two days per week. Each day you provide food enrichment, please only give one treat type. Please **do not exceed the specified amounts** as this can be detrimental to health.

Type	Maximum Amount
 Timothy Hay Cube	1 cube
 Larger Dried Fruit	1 piece
 Larger Dried Veggies	2 pieces
 Raisins	3 pieces
 Small Mixed Fruits/ Nuts/ Veggies	5 pieces

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