

**Nonhuman Primate Behavioral Management Plan
Oregon National Primate Research Center
2020**

A. Overall goals of the ONPRC Behavioral Services Unit

The primary goal of the ONPRC Behavioral Services Unit (BSU) is to provide the nonhuman primates (NHPs) at the Center with conditions that afford them opportunities to express species-typical behaviors and reduce stress, thus producing better research animals. Pursuant to this goal, the primary objectives of the BSU are to provide social opportunities and environmental enrichment promoting species-typical behaviors for the monkeys, to assess and try to decrease abnormal behaviors and to promote animal well-being by using and developing techniques, devices and procedures that contribute to their psychological health.

Staff:

The ONPRC BSU is responsible for carrying out these objectives. Currently, the BSU consists of 12 full time staff. The positions are as follows:

Nonhuman Primate Behaviorist: Head of BSU; responsible for overseeing the unit including the research activities, preparing reports, interacting with Division of Comparative Medicine (DCM) and scientific staff

BSU Manager: responsible for overseeing the animal behavior technicians, interacting with investigators and DCM staff, maintaining records and assisting in the day to day operations of the unit. Also responsible for assisting in the design and implementation of several research projects

Training Specialist: responsible for overseeing the NHP Positive Reinforcement Training program, including teaching others to train, producing shaping plans, and maintaining and analyzing training records.

Social Housing Coordinator: responsible for analyzing data from pair observations and overseeing pair attempts. Also responsible for developing tools for monitoring group dynamics, in an effort to identify factors that could influence group stability, and playing a major role in group formations and harvest selection.

Animal Behavior Technicians (8 FTE): responsible for socializing monkeys (in pairs or groups), monitoring pairs and/or groups for compatibility, providing enrichment and attending to the behavioral needs of the monkeys in their areas.

Interns: The BSU often hosts interns, including summer interns, postdoctoral fellows, graduate students, etc. These interns typically engage in research aimed at improving psychological well being for NHPs.

In addition to the aforementioned duties, each member of the BSU team may be involved in research projects designed to examine psychological well being issues. The BSU works closely with the clinical, husbandry and animal resource staff.

B. Social Enrichment

In the past decade, there has been an increasing awareness of the importance of providing social housing for nonhuman primates (NHP) used in biomedical research. Macaques and other nonhuman primates are social animals and have evolved to live in complex societies. They form intricate social relationships in the wild, where they spend a great deal of time engaged in social behaviors such as grooming and huddling with other members of the troop. Providing social housing for NHPs affords them the opportunity to engage in species-specific behaviors and to develop social and cognitive skills necessary for group living (de Waal 1991). It is commonly postulated that group living provides the best overall psychological enrichment for captive NHPs (Crockett 1998, de Waal 1991, National Research Council 1998). In support of this tenet, studies have reported that socially housed rhesus monkeys display more species normative and fewer abnormal behaviors than singly caged conspecifics (Schapiro et al., 1996, Lilly et al., 1999). Therefore, one goal of the ONPRC is to socially house as many of the NHPs as possible, either in groups or pairs (e.g., Bernstein 1991, Erwin & Sackett 1990, Novak et al., 1992, Reinhardt 1991, Ruppenthal et al., 1991).

B.1 Group-housed animals

Currently, approximately two thirds of the NHPs at ONPRC are housed in groups. Group housing conditions include outdoor corrals (in place since approximately 1972), harem groups (1991), indoor-outdoor “runs” (1964), indoor group “pens” (1998; 2005; 2018), environmentally controlled outdoor shelters (2001; 2005; 2008), and PENs (2013). New facilities for group housing are continuously being planned, funded, and built.

The BSU provides input into selection of animals for social groups, including assessing temperament and compatibility of animals. The BSU participates in the Colony Epidemiology Group (CEG), which consists of representatives from behavioral, clinical, husbandry and animal resources units. This group meets weekly to determine disposition of animals removed from groups (e.g., permanent removal, return to group, etc). BSU also participates in Colony Planning Group (CPG) meetings, in which issues about group stability, project groups, and long-term plans for the breeding colony are discussed. These meetings have greatly improved communication among the various units within DCM, and have helped us to better manage our group-housed animals.

The BSU monitors all social groups on a routine basis, and directs focused attention on groups of concern (i.e., those with a high potential for instability, such as groups with increased aggression, recent changes in the hierarchy, those with a temporarily absent dominant animal, and newly formed groups). The primary goals of routine monitoring of groups include identifying high-ranking individuals and assessing behavioral patterns within the group, in an effort to aid early detection of social instability and facilitates decision making if/when social instability occurs. Information on high-ranking animals may be posted on the outside of group enclosures and/or maintained in PRIME.

Additionally, the BSU monitors transfers from temporary housing back to social groups (“releases”) that are identified during CEG meetings as risky for the individual being released or potentially disruptive for their group. The BSU also monitors groups during times of social instability, and may investigate reasons for the conflict (e.g., by examining records in PRIME, etc).

B.2 Caged monkeys

Since experimental protocols often necessitate keeping animals in cages, one common form of social housing for caged monkeys is pair housing; that is, housing two monkeys in a double cage with a removable slide (which can be replaced to separate the cage-mates). Pairing monkeys allows social contact between the partners and is an acceptable form of social housing. It has been shown that monkeys living in pairs have better immune function than those living in single cages (Schapiro et al., 2000). Our goal for caged animals is to provide them with the maximum amount of social opportunities. Given that continuous social grouping may not always be the best way to meet social needs of macaques in captivity, our plan is to provide some level of social interaction for all primates without creating a situation that would be detrimental to their health and well-being. Following USDA recommendations (APHIS, 1999), we utilize the following methodologies for housing options, in descending order of preference:

1. Pair/Group Housing. Two or more compatible primates of the same species are located in a single enclosure for the majority of their waking periods. This housing is the most preferable, and we place as many primates as possible into pair/group housing.

Pairing of NHPs began with the inception of the Psychological Well-Being Program in 1991. Currently, potential partners are assessed by Behavior Technicians and then moved so that they are next to each other, separated by mesh. Adult pairs are typically separated by mesh for at least 12-24 hours. Juveniles or adults with prior knowledge of each other (e.g., individuals from the same social group) may be separated by mesh for less time, if deemed appropriate by the Behavior Technician. This process allows members of the pair to acclimate to each other. When the Behavior Technician feels the animals have acclimated, he/she slowly removes the mesh slide while observing the responses of each animal. Behavior Technicians might utilize grooming contact and/or clear plastic slides as intermediate steps in the pairing process if deemed appropriate. If there are any signs of overt aggression, the slide is replaced immediately. If there is no aggression, the Behavior Technician closely watches the pair until he or she feels the pair have displayed suitable signs of compatibility (e.g., mounting, tandem threat, sitting in close proximity; McAllister et al., 2020), and then carefully checks on the pair for several weeks following the union. Standardized pair observations are taken on the majority of pair attempts. Husbandry staff are also alerted to the formation of the new pair, so that they can check on the pair. For example, a “new pair” tag may be placed on the cage and a “new pair” sign may be placed on the door to the room, to remind husbandry staff of the newly formed pair. These signs remain on the door for two weeks.

In an attempt to maximize social opportunities for as many NHPs as possible, we may try to pair house a dam/infant (>180 days of age) with a second female. In these cases, the dam/infant pair may be temporarily housed in a cage less than 7.3 square feet (required for a dam < 10 kg and infant < 3 kg) for no longer than 5 days, as the female and dam/infant are acclimating to each other. All animals have adequate space following successful socialization.

If an animal's weight changes with obesity or pregnancy, we may temporarily (usually less than one month, or through the rest of pregnancy) keep the

animal in the cage appropriate for its weight group. This is typically done to allow the animal to remain with its social partner while we try to reduce its weight (or wait for the birth of the infant). Since switching cages and moving animals to new locations can be stressful, allowing animals to stay with their social partner in their original cage can help reduce stress. In all cases, a clinical veterinarian will examine the animal to determine that it is either obese or pregnant, and in all cases, the animal must be able to make normal postural adjustments and movements.

Any problems, including aggression, excessive weight loss by a partner, or other behavioral problems are reported to a BSU technician, who then re-evaluate the pair. Animals permanently separated for incompatibility are introduced to new potential partners as soon as deemed appropriate by the behavior technician. Whenever possible, monkeys in successful pairs are assigned to projects together, to maintain the pair. Behavior technicians are responsible for checking their pairs on a regular basis, to ensure that pairs are not inadvertently broken. Information about pair formation and separations are entered into the electronic health records (e.g., PRIME).

2. Individual housing with visual, auditory, and olfactory contact and tactile interactions. In situations in which continuous full contact is not desired (e.g., when there is concern for the welfare of the animals if there were complete access, or in cases with IACUC-approved scientific justification against continuous contact), continuous availability of grooming-contact bars, which permit tactile interactions between the pair, can be used.

In late 1999, portable grooming-contact cage dividers were developed (after Crockett et al., 1997), which are inserted when a pair of NHPs seems tense or not comfortable as full pairs. Half of the divider is a solid panel while the other half consists of bars wide enough to permit grooming. This design affords individuals privacy and escape from visual contact as well as increased social contact with the cage mate. Animals housed in this situation are considered single housed and therefore this method should only be used in situations in which there is an IACUC-approved scientific justification, clinical justification, or behavioral reason (e.g., animals may be aggressive when housed in full contact but not in protected contact; in this scenario, animals may be housed in protected contact until more compatible partners are found).

3. Individual housing with limited visual, auditory, and olfactory contact but scheduled tactile interactions. In this instance, interactions should occur on a routine, scheduled basis (i.e., daily). Such animals may be housed in a manner that does not permit sight contact with other nonhuman primates, provided that they have scheduled interactions (e.g., be released into a playroom with direct contact with compatible conspecifics). This is not a preferred methodology for housing NHPs, and would require an exemption from the IACUC.
4. No tactile contact with nonhuman primates. In this situation, animals can see others in a bank of individual cages in a room, but direct tactile contact is not permitted. Animals housed in this manner should be provided with additional enrichment, such as mirrors on their cages and daily positive

interactions with human caregivers. The level of additional enrichment will be determined based on factors such as age of the animals, expected length of single housing and behavioral response to single housing. The goal of the Behavior Management Plan is to reduce the number of animals living in this situation by increasing implementation of paired/group housing. Housing monkeys without physical contact with conspecifics should only occur when it is deemed necessary for the health and well-being of the individual monkey (e.g., monkeys with debilitating condition), when there is a documented unavailability of compatible individuals, when there is a documented behavioral reason (e.g., if a monkey is overly aggressive towards other monkeys, or if it appears that the monkey is incompatible with others, e.g., McMillan et al. 2004), or when there is a scientifically justified exemption from the IACUC. Scientifically justified, IACUC approved exemptions from social housing are reviewed by the IACUC annually. Exemptions for clinical and/or behavioral reasons are reviewed by the Head of BSU and the AV every thirty days. A list of exemptions and single-housed NHPs is maintained by the Head of BSU and is available on the Bridge.

5. No contact with nonhuman primates. Unless appropriate justification is made, this system of housing is not in compliance. For example, monkeys that have or are suspected of having a contagious disease must be isolated from healthy animals in the colony as directed by the attending veterinarian. In such cases, the monkey will receive additional enrichment, as approved by the attending veterinarian.

The BSU has developed and implemented a database that tracks all single housed animals, their relevant information (e.g., IACUC assignment, behavioral or clinical issues that may preclude social housing) and their pairing status (i.e., IACUC approved exception, behavioral exception, clinical exception, or in queue to be paired). This report is updated monthly and is maintained on the ONPRC secure network. This information is presented to the IACUC quarterly. The BSU meets with the managers of the husbandry and animal resources unit when needed to help ensure that animals are provided with social housing whenever possible.

C. Nonsocial Environmental Enrichment

A primary goal of our behavioral management plan is to provide monkeys with opportunities to express species-typical behaviors. Thus, we have increased efforts to provide both caged and group-housed monkeys with novel toys and devices that encourage species-typical behavior, such as foraging and exploration. Enrichment devices have been found to decrease the occurrence of some undesirable behaviors, including stereotypies (e.g., Novak et al., 1998) and over-grooming (Schapiro & Bloomsith 1995). The environment of each caged monkey is enriched with cage furniture (e.g., perches) and with a manipulatable object (i.e., toy) that is rotated every two weeks. Novel toys are continually substituted as they are purchased or designed. In addition, caged monkeys are also provided with devices that promote foraging and manipulation (i.e., foraging manipulanda). These devices, most of which are affixed to the cage, are filled with trail mix, grain, and/or produce. Monkeys should receive some sort of foraging opportunity 6-7 days per per week. The toys remain with the cages for sanitation. Husbandry and BSU technicians are responsible for ensuring each caged

NHP has a toy at all times, and for removing and replacing broken items.

Other enrichment includes:

1. Promoting foraging for monkey chow or other food items, including foraging manipulanda.
2. Providing food to the monkeys which utilizes their cognitive and foraging skills, including use of frozen foods (which increases consumption time), placing food on the top of the cage, hiding food (in larger group housing), etc.
3. Providing enrichment for senses other than taste, such as auditory, visual, and tactile, i.e., maneuverable, enrichment devices. For example, the monkeys are given access to television/radio on a rotating schedule. Olfactory enrichment may also be utilized, including aromatherapy.
4. Enriching the structure and substrate to allow species-typical behavior, including substrate through which NHPs may forage.
5. Using novel environments and cages including a “porch” (a small cage that is hung on the outside of the monkey’s home cage, increasing the individual’s visual field of view; Gottlieb et al., 2014) and a “tunnel” (made of stainless steel, it connects two cages, allowing animals to move between the cages).
6. Devices that contain toys as opposed to food, for monkeys on caloric restriction protocols.
7. Cognitive enrichment such as use of tablets or touch screen computers may be utilized (e.g., O’Connor et al., 2015).
8. Increased positive human interaction.

Group-housed animals also receive enrichment designed to encourage species-typical activity patterns. We provide toys that encourage play and exploration, including balls, chew toys and swings. These are checked for wear and tear on a regular basis (e.g., every 2-4 weeks), and repaired or replaced as needed. Play structures provide opportunities for exercise and climbing, and foraging manipulanda encourages foraging. We utilize bedding substrate (wood shavings) in some of our NHP groups (e.g., Doane et al., 2013). Bedding may be used to promote foraging or in times of social unrest.

New types of enrichment items are continuously being developed, tested and implemented. New items are approved by veterinarians and tested on a small number of animals to ensure there are no untoward effects before being implemented across the facility. Such testing typically involves observing animals, either directly or with video, before and after implementation of enrichment. This kind of evaluation can either be performed on a few animals, or may be performed as a “pilot study” in which behavioral outcomes of animals before and after receiving enrichment are compared to behavioral outcomes of control animals (i.e., those that do not receive the enrichment) if deemed appropriate. If, at any point, a problem is noted in an enrichment device, it is immediately evaluated and replaced if necessary. PI staff are notified when novel enrichment devices are introduced.

Enrichment is evaluated on a regular basis.

D. Special considerations

D.1 Infants

Ways to promote psychological well-being for young NHPs include the following:

1. Avoid nursery rearing. Whenever possible, abandoned or orphaned infants are placed with a “foster mother”. Our first choice for foster mothers is lactating females (e.g., females who recently lost their own infant). We have also recently identified several non-lactating foster mothers. These females have been trained to allow infants to bottle feed.
2. Avoid housing infants in single cages with no social contact after weaning whenever possible. The priority is to focus on social housing of young monkeys less than 4 years of age. For infants, this includes “play time” with other infants.
2. Increase weaning age. In 2001, the weaning age was increased from 6 to approximately 8 months of age, in the hopes that a longer time with the mother would reduce aberrant behaviors later on in life. In 2006, the weaning age for indoor housed animals was increased to at least 12 months of age (except for TMB females), as recommended in the IPS International Guidelines for the Acquisition, Care and Breeding of Nonhuman Primates (International Primatological Society, 2006). Infants born in groups are typically kept in their natal groups for 2-4 years unless there are special circumstances. Infants may need to be weaned early for scientific purposes (e.g., maintenance of Expanded SPF colony). In these cases, BSU assesses the infants for temperament and independence. Because highly inhibited infants may be more vulnerable to removal from the dam (e.g., Coleman 2017), infants deemed overly inhibited are typically kept with their dams until at least one year of age.

D.2 Behavior problems

Monkeys displaying undesired behaviors (e.g., self-biting or hair pulling) are reported to the behavioral staff, who then assess and monitor the monkey. Any cases involving injury or wound to the monkey are reported to the veterinary staff. Since 2002, the BSU performs behavior assessments on caged monkeys, to ensure that all animals are observed on a regular basis. Currently, these assessments are conducted twice a year for most caged animals. The behavioral staff assesses monkeys reported as having behavioral problems, in an effort to determine the cause for the problem. Behavior cases are opened for monkeys displaying self-directed aggressive behavior (e.g., Self Injurious Behavior, SIB). Monkeys displaying alopecia are assessed for the amount and location of the hair loss. Cases are opened for monkeys with moderate to severe alopecia (i.e., at least 50% of the body has alopecia). Cases can also be opened for monkeys displaying less hair loss if deemed appropriate by the behavior technician. Information is entered into the electronic animal records.

Behavior technicians try to assess and alleviate the cause of the behavioral problem on an individual basis. One way in which to reduce undesired behavior is to

move the monkey. These moves can include: a) moving timid animals away from aggressive animals; b) moving to an upper level cage; c) moving to a quieter room; d) moving to a room without vocalizing infants; e) moving to a location where more monkeys can be seen, e.g., the interior of a room; f) pairing monkeys if they are not already paired. Another way to ameliorate undesired behavior is with the use of therapeutic devices. These devices include the use of toys to redirect self-biting (e.g., a piece of manzanita wood, rawhide bone, or Kong toy), grooming board or paint roller covered in trail mix, suspended fleece dolls for newly weaned or fearful infants, mirrors for animals to see locations not normally visible (taking care to ensure the monkey receiving the mirror does not respond aggressively to it), and feeder devices (e.g., puzzle feeders, bird feeders, foraging boards). Novel interventions, such as the use of Positive Reinforcement Training (e.g., Coleman & Maier 2010) or porches (e.g., Gottlieb et al., 2014), are also utilized when deemed appropriate.

Monkeys with an open behavior case are assessed regularly; at least every 1-4 weeks, depending on the severity of the behavior problem. New interventions are instituted in non-responsive cases. Data are entered into the animal's history in PRIME using objective criteria.

Because it can be difficult to ameliorate abnormal behaviors once they have begun, in 1998 we began to actively emphasize prevention to avoid the development of these behavioral problems. One of our main preventative measures is to increase social housing, especially for younger monkeys. Other preventative measures include decreasing loud, sudden noises in monkey areas, and decreasing the presence of aversive stimuli such as leather gloves and capture nets around the monkeys. In addition, members of the BSU train new staff on primate behavior, including how to interact with monkeys, and how their actions can affect the monkeys.

E. Training: Operant Conditioning

For several years, ONPRC husbandry staff has been encouraged to provide positive reinforcement (e.g., a food reward) to NHPs for cooperation with techniques such as presenting for injection or vaginal swab or entering a transfer box. Operant Conditioning has been a growing part of our program since 2000. An effective and widely used form of operant conditioning is positive reinforcement training (PRT). With PRT, the trainer reinforces desired behaviors (such as touching a target) by rewarding the animal when it performs the behavior. Training desensitizes the animals to potentially stressful stimuli such as injections (Moseley & Davis 1989, Reinhardt et al. 1990), thereby reducing the stress associated with these stimuli. In addition, by allowing the animals to cooperate with the procedures, PRT gives them some control over their environment (Laule et al., 2003), which can reduce stress (Mineka et al., 1986). Training can also help reduce aggression (e.g., Schapiro et al., 2001) and undesired behaviors such as stereotypical behavior (Coleman & Maier 2010). In 2009, the BSU added a full time NHP Training Specialist position, to help expand and improve upon this training program. A goal is to train as many monkeys (both group housed and caged) as possible to voluntarily cooperate with procedures necessary for husbandry and research protocols, such as entering transfer boxes, presenting a body part for injection, and venipuncture (Coleman et al., 2008). The Training Specialist is also responsible for maintaining training records and assessing and evaluating the training program.

F. Restraint devices

NHPs are maintained in restraint devices (for more than a few minutes) only when required for health reasons, as determined by the attending veterinarian, or for scientifically justified IACUC approved research purposes. In these cases, the monkey is provided with additional enrichment by BSU, husbandry, or scientific staff, including increased human contact, increased manipulanda, or increased cognitive items. Husbandry technicians and others are encouraged to provide positive rewards to NHPs after using short term restraint for procedures such as taking a blood sample or giving an injection. The ONPRC refines restraint devices and/or practices whenever possible. In 2018 we designed and acquired “closed box chairs” (an alternative to the “primate chair” that allows more movement for the animals) and “procedure cages” (a portable cage extender with a reversible squeeze back mechanism which allows procedures such as blood draws to be performed in the animal’s home cage). BSU, husbandry and/or PI staff desensitize animals to these devices.

G. Exemptions from part of Plan

A DCM veterinarian or the Head of BSU may exempt an individual from a specific portion of the Behavioral Management Plan (e.g., food enrichment or socialization) due to health or well-being concerns. The basis of the exemption must be recorded by the veterinarian or BSU Head, and reviewed on a regular basis. The IACUC may exempt an animal from participating in a specific part of the Behavioral Management Plan (e.g., food enrichment or socialization) for scientific reasons. The IACUC must review such scientific exemptions, unless the basis of the exemption is permanent, at least annually. Animals with exemptions from one part of the Behavioral Management Plan still participate in other parts of the Plan (e.g., animals with exemptions from social housing may receive additional enrichment or positive human interaction).

H. Assessing temperament

It has been established that an individual’s temperament can affect response to socialization (e.g., Coleman 2012), enrichment use (O’Connor et al., 2015) and training (e.g., Coleman et al., 2005). Therefore, the BSU assesses temperament as part of the behavioral management program. Temperament is typically assessed cage side, using protocols developed by the BSU. In this temperament assessment, a trained observer not known to the monkey approaches the subject’s cage, standing approximately 1m from the front. Pair housed monkeys are temporarily separated with a protected contact slide (which allows visual and some tactile access between partners) for the duration of the assessment. The observer maintains this position for 5 minutes, taking great care to avoid direct eye contact with the monkey. Using peripheral vision, the observer takes instantaneous focal observations on the subject to record location in the cage (e.g., back, front) and other behaviors of interest (e.g., freeze, stereotypic movement) every 15 seconds. At the end of the 5-minute time period, the observer then makes direct eye contact with the monkey for 2 minutes, again recording behavior every 15 sec. The intruder then presents the subject with various novel objects, one at a time, for 2-3 minutes each. Objects include novel food, brightly colored novel objects, or objects with potentially threatening features (e.g., large eyes, as are placed on Mr. Potato Head). These objects are either hung directly onto the cage or placed on a tray that is temporarily attached to the cage. The observer records the latency to inspect (within 10 cm), intentionally touch, and manipulate (displace from original location) each object.

Overt aggression, urination, defecation, or signs of stress is also recorded. The BSU maintains records on these temperament assessments.

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