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DIRE WOL SPECTES CARE

THE REVIVAL OF A SPECIES FROM ITS LONGSTANDING POPULATION OF ZERO.

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Colossal Dire Wolf Husbandry Manual

This manual serves as a guide to the care and management of the Colossal Dire Wolf. The following Colossal Dire Wolf Standard Operating Procedures (SOPs) are living documents and will adapt and evolve to industry, species, and veterinarian standards as necessary. While it is impossible to cover every scenario in a document, we have developed extensive guidelines to ensure that the best care possible is provided to all dire wolves. As our knowledge of the Colossal Dire Wolf grows, we will continue to advance these standards.

All SOPs listed in the Table of Contents below will be reviewed annually at a minimum and can be updated at any time. Changes can and will be made based on the individual situations, animals, and facilities.

Colossal strives to provide all animals in our charge with the highest standards of care. Colossal challenges our employees and partners to be at the cutting edge of conservation, animal care, genetics, and technology by applying the principles of these fields of knowledge and pushing the boundaries of these disciplines. Our staff does this by carrying the milestones of the past, collaborating with today's change makers, protecting the species inhabiting the Earth, and evolving with the changing needs of all living things. By bringing back species fundamental to our ecosystems, we are creating a better world for the future generations of all species.

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Domestic dogs and wolves do not always have the same requirements. For specific information related to domestic dogs, please see the Surrogate Dog SOPs. If that information does not exist, please refer to the appropriate Colossal Dire Wolf SOPs.

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Background on Dire Wolves

Geography

Dire wolves *(Canis (Aenocyon) dirus)* appeared first in the midcontinental U.S. between 300,000 and 12,000 years ago. They were found as far north as Alaska and down into southern Mexico. Thousands of dire wolf fossils have been collected from Rancho La Brea in Los Angeles, California. In South America, fossils of dire wolves dating to about 17,000 years ago have been found in Venezuela, Peru, and Bolivia. The distribution of the dire wolf suggests it was adapted for multiple habitats, from boreal grasslands, coastal open woodlands, to tropical wetlands.

Genealogy

An eastern subspecies (*Canis (Aenocyon) dirus dirus)* appeared first and was larger. A western subspecies (*Canis (Aenocyon) dirus guildayi*) was smaller and evolved slightly more recently. Along with mammoths and mastodons, the dire wolf died off between 10-16,000 years ago. The timber wolf or grey wolf, developed in northern Eurasia, and probably crossed over the Bering Strait land bridge into North America. The two species, (*Canis lupus*) and (*Canis (Aenocyon) dirus*), coexisted for 400,000 years in North America. However, a recent study looking at the genome of the dire wolf suggests that they are not cousins as previously assumed, but that the closest living relatives of the (*Canis (Aenocyon) dirus*) are the African jackals. This would suggest that the species should be removed from the (*Canis)* genus and would become Aenocyon dirus.

Extinction

Before the arrival of humans, wolves, and dogs, the only competition for the dire wolf was probably from the saber-toothed cats. Their struggle to adapt later on was likely a factor in their extinction. According to Angela Perri, an assistant professor of anthropology at Texas A&M University, "They were hanging out by themselves for a very long time, and specialized in the way they do things. And then modern humans come in and they can do it all."

Size

(Canis (Aenocyon) dirus) looked much like our modern day wolves, only heavier and with larger jaws and teeth. They had relatively shorter legs, as well as larger shoulder blades and pelvic bones. The timber wolf or grey wolf *(Canis lupus)* of today is leaner, usually weighing between 66 and 145 pounds (29.4 -65.8 kg), whereas dire wolves weighed between 125 and 175 pounds (56.7 - 79.4 kg).

Diet

Dire wolves were carnivorous. It is believed that they traveled in packs, hunting large prey like ancient horses, bison, and maybe even young mammoths and mastodons.

Behavior

The large number of dire wolf fossils found at Rancho La Brea have suggested that like modern grey wolves and other canids, dire wolves formed large social groups to help hunt and raise pups. Sexual dimorphism is seen in male and female dire wolves with the males being larger and a bit more robust than the females.

Developmental Milestones of the Colossal Dire Wolf (Based on Grey Wolf Data)

0-1 Week

- Average weight for a grey wolf pup is 0.5-1 pounds (birth weight of the Colossal dire wolf will be compared to this to determine expected growth following birth)
- Blind with closed eyes
- Deaf with small ears
- Pugged nose, with little sense of smell
- Good sense of taste, touch, and has a sense of balance
- Completely dependent on mother for feeding and care
- Limited mobility, utilizing front legs more than hind legs
- Vocalizations include whines and yelps
- Pups should gain an average of 2.6-3.3 pounds a week for the next 14 weeks

1-3 Weeks

- Eyes and ear canals begin to open between day 11-15
- Eyes are blue and not fully developed
- Begin to crawl around and explore their den area
- Continue with gradual weight gain
- Teeth begin to emerge
- Will start to stand and walk
- Vocalizations include whimpers, squeaks, and first howling attempts
- Pups should be around 3.5 pounds between 2-3 weeks

3-6 Weeks

- Begin to explore and venture beyond the den area
- Begin eating regurgitated food brought to them by the mother
- Hearing improves at four weeks
- Ears become erect at five weeks
- Adult hair starts to appear
- Social bonds with littermates develop
- Continue to gain weight and grow rapidly

7-12 Weeks

- Show signs of independence
- Milk teeth develop
- Begin to consume solid food
- Increased mobility and play
- Early stages of modeling parental behavior to learn hunting skills

• Begin weaning from mother's milk

12-16 Weeks

- Weaning completes, pups still eat regurgitated food
- Eyes start to change from blue to amber-gold
- Weight should be around 22-30 pounds

3-6 Months: Juvenile Stage

- Continued steady growth and development
- Learning social dynamics and pack hierarchy
- Refining hunting and survival skills
- Pups gain 1.3 pounds a week for next three months
- Adult teeth come in
- Dire wolf should weigh up to 70 pounds

6-18 Months: Sub-adult Stage

- Nearly adult size and weight
- Full adult coat development
- Increasing independence
- Potential to disperse, seek out mates, and establish a territory

2 Years +: Adulthood

- Fully grown and sexually mature
- Active participation in pack dynamics
- Potential breeding and reproduction
- Territory defense and social interactions
- Hunting and providing for the pack
- By three years the skeleton is fully grown, and adult weight is reached

Radio Communication

Staff should have two ways to communicate within the facility. Suggestions for best practices to ensure safety of all people and animals are "push to talk" radios and cell phones and communication apps for phones such as WAVE; but other manners of communication could be applied. It is recommended to have a backup communication system in case of emergency or if one system is disabled for any reason such as weather or power outages.

Communication among the staff is paramount to the safety and wellbeing of the animals, staff, and partner facilities. The push to talk radios that are used by staff allow information to be shared easily and effectively throughout the regular workday as well as in an emergency. Facilities are responsible for teaching staff how to work the radios so that radio transmissions are clear and audible, modeling and monitoring appropriate use, and reminding staff about proper language and use of radios as necessary. It is the responsibility of staff members to use radios correctly and ask for guidance if needed.

Radio Language

Radio transmissions are intended for sharing information that is vital for the safe and efficient operation of the facility. Radios are not to be treated as toys or opportunities to be "on the air." The language used should be free of profanities, clearly spoken, and convey an understandable and audible message or question.

It is expected that all radio users be mindful of the language that is used on radios as transmissions are accessible to local law enforcement and news agencies, and guests that may be in presence of staff.

Radios must be turned on, volume set to an audible level, and carried during the work hours. To make a radio call:

- Please take a moment to think about what you plan to say on the radio.
- Who are you contacting?
- What is the information you need to convey or question to ask?
- Wait ~ three seconds before speaking (radios have a short delay; your voice may be cut off if you speak immediately).
- Holding the radio no closer than six inches from your face; speak to the radio with a clear voice.
- Say "your name" to "other person's name," then release the button and wait for the other person's response –ie, "Steve to Matt."
- Depress and hold the button, wait approximately three seconds, and speak clearly to answer.
- Repeat as needed.

Habitat and Enclosure Requirements and Descriptions

This document provides details of size, complexity of habitats, holding area, training facilities, natural/ manufactured bodies of water, types of fencing, types of shade (natural vs. synthetic), how animals can be moved around within the entirety of their facility, and number and location of gates for security checks. All gates and locks should be checked daily regardless of use.

Colossal requires that all animal habitats, holding, dens, den boxes, etc. have cameras set up to monitor animal behaviors. The Head of Animal Husbandry and Manager of Animal Husbandry will have access to the cameras and will ensure that all responsible parties have access to the camera systems. Cameras should have night vision, recording, and zoom features. Every effort should be made to have as much of the habitat covered in the camera or the cameras so they can be remotely moved to see all areas.

Outdoor enclosure size will be a large environment encompassing many natural features and topography. This will allow Colossal dire wolves to exhibit natural behaviors. This will include natural substrate (dirt, grass, sand, leaf litter) with a varying topography. Furnishings within the area will include rocks, logs, tall grasses, shrubs and bushes, and trees. By adding more plantings, the habitat will provide natural visual barriers where dire wolves can seek refuge behind/in/under. Dire wolves may seek to dig and create their own dens in the habitat, but careful consideration should be given as to whether an artificial denning area would be appropriate to provide. These decisions should be made on a case-by-case basis and take into consideration the individual animal, the pack dynamic, and facility.

Containment barriers will be sufficient to prevent the dire wolves from leaving their enclosures. Some individual dire wolves may be great jumpers and climbers. The following containment barrier is recommended:

- Wire fencing will be of appropriate strength, with support posts solidly anchored in the ground. A nine gauge or heavier two-inch chain link suspended on at least two-inch steel pipes that have been set in concrete is recommended.
- Vertical height will be eight feet or higher, with a 36-inch overhang at a 35-45 degree angle.
- Vertical barriers will include an underground component that is four feet inward from the main fence to prevent dire wolves from digging out.
- All primary containment barriers also have several strands of electrical hotwire as a further deterrent from testing the fence. Hot wire should not be used as primary containment as a dire wolf will run or jump through it if stressed or startled.
- The entire enclosure is also surrounded by a secondary fence of the same construction including height, hotwire, and dig barriers providing redundancy and a secondary containment option for further containment of dire wolves. Smaller containment areas may need to have the entire top covered as the overhangs may not be sufficient for containment.

The Association of Zoos and Aquariums (AZA) and U.S. Fish and Wildlife Service (FWS) recommended minimum size requirements:

• Three same-sex or non-reproductive dire wolves should have a minimum 6,000 square feet habitat and three holding/shift pens at 200 square feet each.

- A single generation breeding group should have a habitat of no less than 10,000 square feet, with a minimum of three holding/shift pens at 200 square feet each.
- A multi-generation breeding group should have a main habitat of no less than 10,000 square feet, a secondary habitat at 5,000 square feet, and at least three holding/shift areas of 200 square feet each.

Habitats and holding areas should be big enough that pack members can move about and allow enough space to support pack hierarchy. Large areas with minimal corners and no less than a 90-degree corner should be in the habitat. Multiple visual barriers should be provided to help lessen competition and make subordinate members of the pack more comfortable.

The habitat should provide flat areas where dire wolves can walk and run. Slopes or hills of dirt and soil are helpful in creating more dynamic topography. Plants, deadfall, log furniture, human made dens and shelters should be provided. Dire wolves are great diggers, so dig barriers should be in place around the habitat's perimeter. Sufficient shelters included in outdoor enclosures allow for them to choose to seek protection from the elements, as well as insects, and to seek privacy from one another or human disruption if desired.

Human-made dens are better for monitoring dire wolf activity and ensuring pups are thriving. Human-made dens constructed out of wood, block, concrete or PVC can be provided to try and help prevent dire wolves from digging their own dens in the habitat. Dire wolf-made dens can collapse, so monitoring and treating individuals can be a significant challenge. Dire wolf-made dens should be filled in daily if the animal husbandry team determines they don't want dire wolves to utilize them.

- Dens should be large enough for a female to lie down with legs extended, as she would when nursing.
- The den should be high enough for the dire wolf to stand upright, but not much higher than that.
- Smaller dens can be provided for warmth and to give the dire wolf choice. The den box should be large enough for one dire wolf so a nursing mom can't lie too far away from her pups.
- There should be multiple dens provided year-round. Every dire wolf should have a den and some institutions provide a few more so subordinate pack members can have a den and not be displaced.
- The substrate in the den should have a wood floor, making sure no places for toes or toenails can get caught. The wood should be covered with straw, hay, or shavings (consideration should be made to individual behavior).
- Don't use materials that can be ingested such as blankets, wood chips, or domestic dog beds.
- Careful consideration should be given to den substrate when pups are forthcoming or present. Pups can get sores on their pads due to the wood floors or harder substrates—good options are dirt or soil.

In addition to large outdoor habitats, smaller holding spaces will be used for the purposes of off habitat holding, shifting, and isolation for social or medical reasons. Regular access will be given to these areas to ensure that wolves are familiar and comfortable with going into these areas. These areas can make capture much easier. Feeding a portion of the animal's diet regularly in these areas can help animals to feel comfortable going into the smaller holding areas. At least some of these areas should be climate controlled to allow for specialized care when needed.

Dire wolves are naturally shy and prefer to stay out of the direct open. Natural plantings of assorted sizes should be planted to help provide privacy for the dire wolf and give a sense of security. If dire wolves don't feel secure, maladaptive behaviors can develop. Stereotypic behaviors can include:

- Pacing
- Spinning/twirling

- Over grooming including licking and chewing of self, pups and conspecifics
- Aggression
- Diarrhea
- Hair loss
- Self-mutilation
- Maternal neglect
- Loss of appetite
- Weight loss
- Isolation from pack
- Jumping/scaling fences
- Disease

Design of habitats and enclosures allow for large machinery to access the areas to assist with adding, maintaining, and removing substrates, trees, den boxes, and other large furniture. Vehicles may need to be used to maintain the security of both the interior and perimeter fencing. All facilities should have an access road between the primary and perimeter fences that allow for these areas to be carefully checked daily.

Environmental Parameters

According to the AZA Mexican Gray Wolf Husbandry Manual, the Mexican grey wolf has been housed from Texas to Michigan successfully without environmental heating or cooling. Where the animals have adapted to extreme ranges in climate and temperatures, Colossal requires that we provide the Colossal dire wolf with choice and control in their managed environments. Heating and cooling such as air conditioning or evaporative cooling should be available to geriatric, ill, young and other individuals or situations as deemed appropriate by the Head of Animal Husbandry and veterinarians. Heating should be provided to geriatric, neonates being hand reared, and animals with compromised health if temperatures fall below 45° F.

A healthy adult dire wolf is cold weather tolerant. They must have dry dens with dry bedding, wind breaks, and multiple shelters/dry areas for animals to get out of cold and wet conditions. If temperatures are below freezing (32° F), supplemental heat should be provided in dens and indoor holding. Care should be given in making sure there are enough dry and warm places for all members of the pack to seek refuge in. When supplemental heat needs to be supplied, make sure that carbon monoxide detectors, fire detectors, and fire extinguishers are nearby, and all safety measures are taken to prevent burns to animals and to prevent the straw or other bedding materials from catching fire.

During the hotter months, adult dire wolves should be provided with plenty of shade, misters, and water features where they can lay in and cool off. In extreme heat, air conditioning and evaporative coolers can be used to help dire wolves cool down. Dire wolves pant to cool themselves. If it is hot and they are not panting, this could be a health indicator. Please contact your supervisors and let the Colossal husbandry team know. Cooling rocks and cooling pads are good options to bring the dire wolves to locations that assist in their husbandry and care. Ice treats and frozen blood treats can assist in cooling animals down and provide enrichment and feeding opportunities.

Pups three weeks and younger can't thermoregulate and special care should be taken to keep them in a temperature-controlled setting, so they don't overheat or get too cold.

- The pup(s) environment should be kept at 85° F until the pup is one month old if being hand reared, and depending on its health its environment, may be kept at this temperature for longer.
- If the pup(s) are being reared by their dam, temperature should be kept between 65-75° F and should never be below 45° F for neonates.
- If the pup seems cold or lethargic, animal care technicians should contact their supervisor. The dam should help to provide the correct temperatures but building and floors can be controlled to assist the dam and pack in creating the correct environment and can prevent the pack from moving pups, or digging deep dens.
- A pup's rectal temperature should be between 100-101° F.

• If a pup needs to be warmed up, wrap the pup's body (never cover the pup's head) in warm dry towels, use a heat lamp, heating pad, or a hair dryer. Be mindful not to burn the pup as they cannot regulate their temperature and will not be able to move away from the heating device.

Light cycles are important to dire wolves as they are seasonally reproductive. The season is triggered by the daylight cycles. The alpha female's estrus is also triggered by light cycles, so keeping the indoor light cycles the same as the outdoor natural cycles of the region they are housed in is helpful for breeding dire wolves. Timers with full spectrum lighting can be used in indoor housing areas and should be set to mimic the light cycles occurring in the Northern U.S. Light cycles also play a significant role in the shedding cycles of both male and female dire wolves.

Dire wolves have access to multiple water bowls, ponds, and/or pools. Water quality is scheduled to be tested regularly. When dire wolves have pups, ponds/pools and water bowls/troughs should be lowered to a level to ensure the safety of the pups. The pups should be able to get out of the water features without having to swim. The water should be at a level where the pup can stand with their head above water.

A pup should not be trapped in a water feature, where if the pup gets tired or can't get itself out, it can rest its legs and head without the water level putting the pup in danger. Pup-proofing the holding and enclosure should be done before the pups' arrival. Pup-proofing should include but is not limited to filling, blocking, and modifying large gaps, spaces between gates, fences, furniture, dens, and any structures the pup would have access to. Areas and spaces in and around fences, gates, dens, night houses, barns, rock, deadfall, and climbing structures should be filled with solid barriers. Smaller gauge fencing can prevent escape, getting stuck, and injuries to pups.

Consideration to sound levels near the dire wolves should be monitored to make sure it stays within safe levels as dire wolves have good hearing and loud noises can damage their hearing and get in the way of their pack communication.

More information regarding extreme temperatures such as extreme arctic/winter temperatures can be found in the <u>Emergency Response section</u>. Information regarding the temperatures for Colossal dire wolf neonates can be found in the <u>Neonate Rearing Guidelines</u>. If any questions arise, please contact the Head of Animal Husbandry for further clarification. This is imperative to ensure the Colossal dire wolf receives the best welfare possible.

Protected and Free Contact

Free contact is when humans and an animal share the same unrestricted space. There are no barriers between the human and the animal.

Protected contact is when an animal and human do not share unrestricted space, and some type of protective barrier is used. The animal is not confined or forced to interact and can choose to leave the work area at will.

There is no free contact with the Colossal dire wolves, not including domestic dogs over 45 pounds unless approved by the Head of Animal Husbandry. If an emergency situation arises where it becomes necessary to enter a space shared by a Colossal dire wolf, the Colossal Head of Animal Husbandry will give additional guidance based on the situation.

If free contact is ever deemed necessary, the following will need to be considered:

- One more staff member than there are wolves is required.
- No food of any kind can be brought in, including gum, human candy/food, dry dog treats, etc.
- Make sure there is no dried meat or other food items on your clothes or shoes. Change if needed.
- Make sure you do not have extra items on you or on your belt loop that the wolves could jump up and latch on to (e.g., sunglasses, toys, etc.).
- Pepper spray is to be worn, and your radio must be on your person.
- You must carry a rake, shovel, net, or y-pole.
- Make sure the wolves and your coworkers always have your undivided attention when in with the wolves.
- Do not use cell phones or smart watches while in with the wolves unless it is an emergency.
- Do not let the wolves get near you, do not crouch down, do not attempt to play with the wolves, ignore bad behavior and reward good behavior.
- Remove sunglasses as this may adversely affect the animal if they are not used to seeing humans in sunglasses. Facilities and staff may choose to habituate the dire wolves to seeing people in sunglasses.

Behaviors to be trained will be prioritized with the animal management team. Decisions will be made considering the needs of the pack, the individual, habitat, and resources available. Supervisors and the Head of Animal Husbandry will determine which behaviors, frequency of training, and shaping plans will be utilized. Training will be done in a protected contact setting unless it can be determined and approved by the Head of Animal Husbandry that is completely necessary to reach the goal behaviors, and it is safe for the staff and the animals. Time and ease of training the behavior or utilizing free or protected contact will not be considered necessary unless it is a quality of life concern. Free contact comes with significant risks to human safety and can cause animal safety concerns and animal escape concerns. Free contact may be utilized for special cases such as hand rearing and will be outlined in the species-specific hand rearing SOPs. Most nondomestic animals (usually everything except domestic dogs and cats) prefer protected contact as it gives them control of their environment and allows them more choice, often providing better welfare options for the animal.

Servicing Standards

Colossal takes great pride in providing the highest standard of animal care to Colossal animals. Standards should be at the very minimum AZA standard level and above. This includes providing the best habitats, environments, and diets to ensure the highest levels of welfare. If the animals' areas are dirty, soiled, or pride is not put into caring for the habitat and holding areas, this can create small impacts on animal welfare and combine into larger problems over time.

Colossal also understands that excellent welfare does not always equate to an immaculately clean area. Animal care technicians, managers, and facilities personnel need to be mindful that maintaining habitats and holding areas that are overly sterile does not provide for a dynamic, complex, and naturalistic environment. Some areas may be scent or territorial marking and may be left to help the dire wolves communicate and mark territory. Trees, wood logs, furniture, and substrates don't need to be disinfected as it will be hard to effectively clean natural items, but feces must be removed once a day.

Scent plays a very important role for dire wolf communication and over cleaning and sanitizing may negatively impact the wolf's psychological welfare by not allowing the individual or pack to have a secure territory. As olfaction is an extremely important aspect of a wolf's senses in how it interacts with its environment, care should be taken to allow for this to be expressed through scent marking and inspection of scent areas. Finding the balance is the key to ensuring Colossal dire wolves have the highest standard of care possible.

Colossal husbandry team will work together to find best practices that ensure the highest standards of welfare and sanitation at the dire wolf facility.

Disinfecting shoe baths, also known as footbaths, should be used at the entrance to the buildings or areas housing any Colossal animals. This best practice will help prevent the spread of pathogens to Colossal animals.

When servicing enclosures, habitats, night houses, dens and holding yards, care should be taken to provide good sanitation of all areas to reduce the instances of intestinal parasites, pest species, and disease.

- Removal of uneaten food, bones, and feces will help prevent attracting insects and other pests to the area.
- Hard surfaces should be cleaned daily and thoroughly disinfected weekly unless directed by veterinarians or senior staff as sometimes elimination of scents can cause stress to individuals.

• Basic cleaning and removal of feces should occur daily from holding areas, and at least weekly from larger habitat areas.

Animal caretakers should pay attention to signs of pest species such as insects, feral cats, feral dogs, skunks, raccoons, foxes, bobcats, etc. These animals pose health risks to dire wolves. Feral and wild animals can serve as vectors for many diseases and parasites such as rabies, leptospirosis, parvovirus, canine distemper, and toxoplasmosis. When using pest control measures for rodents and other species, care should be taken to ensure that dire wolves will not have access to, consume, or interact with animals that have ingested poisons or rodenticides. The Head of Animal Husbandry and the veterinarian should be consulted before employing any pest control plans. Pest control measures can be found in the dire wolf <u>Preventative Medicine Program</u>.

Animal care staff should clean animal areas daily, including but not limited to water bowls, feed bowls, cement surfaces, and feces, urine, or other bodily fluids. The type of substrate and materials of the exhibit and holding yards will determine how areas are cleaned. Colossal dire wolves' food and water bowls should be cleaned with soap and water daily and sanitized 1-2 times a week or more often if needed to eliminate food borne pathogens. Food debris should not be left in the exhibit for over 24 hours.

Substrate areas should be raked of debris and any latrine areas should have substrate changed by digging out old, saturated substrates and replacing with new substrate when area becomes overused and natural decomposition is no longer occurring due to the build up of urine. This will vary depending on the location and the weather, as well as the number of animals and size of the habitat.

Soiled bedding should be removed daily and replaced with fresh and clean bedding, unless a dam has pups or is getting ready to deliver pups. Den changes can cause the dam to move pups, increase stress on the dam and pups, or cause the dam to injure or abandon pups altogether. Once the dam starts preparing her den to deliver pups, bedding changes should be kept to a minimum and only done opportunistically after risk assessment of the situation. Cement holding areas should have debris removed, and then cleaned and disinfected per established schedules. Squeegees should be used to help dry the floors to prevent slip hazards, mold and mildew, and algae from growing. Bedding should be removed and returned if still usable or replaced if soiled with feces, urine, or smells of mold and/or mildew.

All animal waste should be disposed of in accordance with state, federal, and species regulations. Feces should be bagged and tossed into the appropriate receptacle.

If the containment structure includes windows, they must be cleaned regularly with a non-toxic cleaner or a combination of vinegar and water. Chemicals used should be approved by the Head of Animal Husbandry.

Pools or large troughs should be drained and bleached at least once per week. At no time should a pool or trough have slimy sides/bottoms. They should not have debris (a few leaves/grass flakes are fine, but handfuls are not), appear to be green, or have algae starting to grow or be present. If you wouldn't want to drink the water, the animal should not have to drink the water. Periodic water testing, at a frequency of no less than once a year, should be done to ensure the drinking water is safe and free from pathogens.

Ponds or large tanks without drains should be monitored no less than seasonally to ensure that the water quality is appropriate as the dire wolves will lay, swim, and drink the water. If water quality is not human grade or safe for animals to drink, the water should be treated at the direction of the Head of Animal Husbandry and veterinarian.

Enrichment items should be cleaned after every use, so they are available and ready for the next use. Plastic/ metal and other cleanable enrichment should be washed with soap and water after each use. If the item is used for meat or prey items, the enrichment device should be disinfected after use. Once items are dry, they should be stored in the appropriate location.

Animal care technicians should remember to clean walls, beds, shelves, and areas where dire wolves may drag or carry food and rub up against. If these areas are contaminated with food, urine, or feces, clean them with soap and water. Any plastic, metal, cement, or other disinfectable areas should be cleaned with soap and disinfected in accordance with the <u>Disinfection and Cleaning SOP</u>.

All cleaning, disinfecting, and servicing will be recorded in the animals' daily record in the animal care information management system.

For more details, please see the following sections:

- <u>Preventative Medicine Program</u>
- Disinfection and Cleaning
- Behavioral Husbandry Policies and Procedures
- <u>Record Requirements</u>

Shifting Procedures and Precautions

When shifting Colossal dire wolves, positive reinforcement measures should be used. At no time during shifting, should animal care technicians be in a space that a dire wolf over 45 pounds has access to. The Colossal dire wolf is a large and powerful predator and should be given the respect that accompanies working with large carnivores and omnivores.

Shift and recall training should be utilized to ensure the dire wolf can be brought into and secured in the holding facilities in case of weather, emergencies, conspecific aggression, or unforeseen occurrences that could cause safety or security issues. This can also be utilized should a non-inventory animal land in the habitat or a non-approved food item find its way into the exhibit.

Shifting should be done daily and become a part of the wolf's routine. Staff should be calm and patient when shutting doors and securing animals in normal situations. On occasion, animals may go back and forth or not be clear of the door. In these instances, staff will need to practice patience and either give themselves and the dire wolf a timeout and regroup after a short time period or attempt a different reward or method of shifting. If animal care technicians become too frustrated, they should communicate the issue to the supervisor to assist in troubleshooting the reason for the shifting challenge. When necessary, additional staff may be required for successful shifting. Please see <u>Behavioral Husbandry Policies and Procedures</u> for more details and contact the Head of Animal Husbandry with any questions.

When shutting and securing doors, animal care technicians should be mindful not to slam shut doors on any part of the animal as this can cause severe injury and create a situation where the dire wolf will no longer shift or feel comfortable in holding areas. Guillotine doors should never be released before the entire animal is completely cleared of the door. Shutting a door on the back of a dire wolf can create permanent spinal injuries to the animal. Slide doors can cause injury to tails, legs, eyes, ears, and bruise animals' torsos. Swing doors can also injure both animal and care staff if they are not used properly. Animal care technicians should be present and paying attention to the dire wolf and not distracted when shifting. They should not be on their phones, radios, or talking to their team members when actively opening and closing doors. A dire wolf should not be caught or trapped in a space unless it is directed by a supervisor and/or an emergency. In this situation, the animal care technicians should have the direction of the animal care supervisor. Animal care staff should also be mindful to not put hands/feet in mesh/gaps or areas where the dire wolf can bite or scratch them.

For more details, please see the following sections:

- Behavioral Husbandry Policies and Procedures
- Protected and Free Contact
- Lock Out Tag Out (LOTO)

Daily Health and Wellness Evaluations

Animal care technicians are a vital part and often the best advocate for noticing when an animal's health or wellness may be compromised. They are expected and required to observe and report any changes in the animals' physical or mental well being. When reporting and documenting changes, it is important that animal care technicians document changes in clear and defined terms. The animal care technician should analyze what is off and record and communicate that in the appropriate manner including the daily report and if warranted, notifying their supervisors. Is the animal eating normally, walking normally, interacting socially in its normal manner?

If the animal care technician is new to working with the animal, they should seek out the advice of a person familiar with the individual or the species. An individual animal's idiosyncrasies should be notated in the animal's record and be a part of the animal care technician training program so that new staffers are aware of any issues and can monitor for progression or regression. Animal care technicians spend the most time with the individual animals and should also be mindful of the blindspot that this can create by not noticing subtle changes that occur over long periods of time. This can be mitigated by use of animal care technicians that don't work with the animal every day but are working with them weekly. Supervisors and veterinarians will also play a role in noticing subtle changes in the animals. It takes an entire team of animal care technicians to ensure that the animals are receiving the best care possible.

Individual animals should be checked from the tip of their nose/ears to the tip of their tail/toes at least once daily to monitor overall wellness.

Check for:

- Wounds
- Bleeding
- Limping
- Unusual swellings or discharges
- Changes in weight or appetite
- Diarrhea or abnormal feces
- Inability to defecate
- Labored breathing
- Any abnormal behavior such as increased aggression or lethargy
- Fly bites on/in the ears

Other daily observations:

- Dietary intake
- Fecal quality (Regular fecal checks should be done to monitor individuals for intestinal parasites and diseases. Fecal scoring should be used, and staff should ensure a common communication method amongst care takers.)
- Body condition scoring
- Coat condition
- Activity level
- Social behavior and age

Wolves should be monitored for interactions with conspecifics, allospecifics, and their animal care technicians. Changes should be noted in the animals' records and communicated to the supervisors and other animal care technicians in charge of the animal.

The following example is from the AZA Large Canid Care Manual:

Table 11: Gastrointestinal grading sheet for red wolves (adapted from Waddell 1998)

Grade 1	Greater than 2/3 of feces in a defecation are liquid. Feces have lost all form, appearing as a puddle or squirt.
Grade 2	Soft/liquid feces are an intermediate between soft and liquid feces, with equal amounts of soft and liquid feces within the defecation.
Grade 3	Greater than 2/3 of feces in a defecation are soft. The feces keep enough form to pile, but have lost their firm cylindrical appearance.
Grade 4	Firm/soft feces are an intermediate between the grades of firm and soft, with equal amounts of firm and soft feces within the defecation.
Grade 5	Greater than 2/3 of feces in a defecation are firm. They have a cylindrical shape with little flattening.

Body Condition Scoring

Body Condition Scoring (BCS) should be used to help decide an animal's overall condition. Body Condition Scoring can be used with weights, but animal care technicians should be mindful of an individual's age, health condition, hierarchy, reproductive cycles, and bone and body structure when employing weights as a measure of body condition. Body Condition Scoring systems should be done often and by trained individuals. The Nutrition Advisory Group to the Association of Zoos and Aquariums lists the following tips on how to body score animals. Body Scoring is a subjective tool and one of many that can be used to monitor the health and wellness of Colossal dire wolves.

Body Condition Scoring is best learned with consistent practice. It can be helpful to have multiple staff members familiar with the species involved at the start to ensure that staff is consistent in the way they approach and look at the animals. This helps everyone use and be familiar with terminology, anatomy, and the scoring system. As the team becomes more skilled at using BCS, it may be beneficial to limit the number of scorers making sure to include a person who doesn't see the animal every day to help notice variations.

Scorers should be familiar with the anatomy of the species. If multiple people are used to assess and give BCS, they should be regularly and objectively evaluated for consistency with the same animals. Recording the initials of the scorer can assist in following up for clarity and tracking trends. Colossal dire wolves will be scored with the nine-point scoring system and no half scores will be given. This allows for scorers to become familiar and proficient with the scoring system. Having the same system in place allows for trends to be captured in individual animals and the species.

A schedule noted in the animal care information management system should be adhered to and will ensure regular and consistent BCS is taken. BCS can complement weights and can be used for animals that are not weight trained or are reluctant to step on the scale. Colossal dire wolves will be trained for voluntary weights as soon as reasonably possible. Per the <u>General Communication Guidelines</u>, Body Condition Scoring will be done at a minimum of every month at visual inspections by the attending veterinarian, area supervisor, and appropriate animal care technician. The Colossal husbandry team will do BCS on all Colossal dire wolves every time they visit a site which will be no less than twice a year. Please see the <u>General Communication Guidelines</u> for more details.

For pups, juveniles, and pregnant, nursing, and post pregnant animals, body weights coupled with the aid of a BCS system will assist scorers to adjust for growth curves and development. It assists in identifying how the pups and pack are doing and making sure their growth is happening at an appropriate rate.

BCS can help identify the target weights for individuals for the season and life stage they are at.

See Body Condition System Chart or follow the link: Canine Body Condition Chart



Advancing Science for Pet Health

BODY CONDITION SYSTEM



The BODY CONDITION SYSTEM was developed at the Nostlé Parina Pet Care Center and has been validated as documented in the following publications Lalarma DP. Devlopment and Vikidition of a Redy Candion Scare System for Days. Cartier Pravitic Adj, August 2007 Radow DR et al. Comparison of Vision Middle for Estimating Body Frist Days. Also Aithe Roy Assoc 2002, et 2: 309–44. on of Vari

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Daily Husbandry Routine Outline

Opening Colossal Dire Wolf

- Check email and read reports from the previous day.
- Check notes for any animal(s) that might require medication.
- Check weather and follow <u>Emergency Response SOP</u> and the facilities' specific inclement weather SOP if needed.
- Take the assigned radio and bear spray.
- Make sure all appropriate lights are on and functioning correctly.
- Conduct life checks
 - Make sure all animals in the collection are alive and well. Initial check off list must be done within the first hour of your shift.
 - After getting a positive count on animals locked inside, follow any <u>Lock Out Tag Out (LOTO)</u> <u>SOP</u> to enter the exhibit space.
- Physically and visually check integrity of all exhibits for any breaches or escape hazards in exhibit; including but not limited to fencing/mesh/support poles, electric fence and insulators, fallen trees, digging along fencing, and other miscellaneous structures. Initial check off list when completed.
 - Test hotwire: hotwires should always read at least 5V or higher. Anything lower, inspect for any grounded-out areas. If an issue cannot be seen or resolved, contact the senior most animal supervisor or facilities manager. Record reading on check-off list in the animal care information management system.
 - Physically double check that all exhibit access points are locked and secured. Many exhibits have multiple points of entry. Make sure those that are not used frequently are checked and locked daily.
 - If any breaches are found, notify the lead conservationist immediately and do not let animals on exhibit until repaired.
 - Remove branches, debris, snow from covered exhibit tops.
- Check integrity of secondary fencing/hotwire surrounding the exhibits. Initial check off list when completed.
 - Physically make sure any gates/access points are locked and secured.
 - Pick up garbage around the outside of exhibits.
- Check weekly task lists for full cleans.
 - All cleaning may be completed in any order most convenient to the animal care technicians for that given day.
 - Change all footbaths daily and when they have more than 10% debris in them.
 - Follow <u>LOTO SOP</u> for entering habitats.
 - Water bowls/feeders/food enrichment should be cleaned with soap daily and disinfected at minimum once per week.
 - Clean up exhibit, remove animal waste, soiled straw and bedding, and non-collection animal waste. Dispose of waste in a bag in the proper receptacle.
 - Clean windows.
- Check exhibit pools. Drop, scrub, and refill as needed. Turn on any waterfalls as needed.
- Put necessary food items out in the exhibit for the Colossal dire wolves. Please check with the Head of Animal Husbandry for a list of currently approved diet and enrichment items for the specific individual.

- Check notes for any animals that might require medication.
- If venomous snakes and other dangerous animals are present in the area, areas including but not limited to dens, night houses, barns, exhibit deadfall, and climbing structures should be visually checked for venomous snakes and the venomous snake removed by a staff member trained in removal of venomous snakes or other dangerous animals. Snake bites are common in dire wolves.
- Provide any enrichment/training for the day. Hold back part of the diet if needed for training or medication later in the day.
- Be sure to take out everything you brought in!
- Check exhibits to make sure any additional personnel are not in there.
- Lock/secure truck gates and exhibit access doors.
- Double check exhibit locks.
- Remove LOTO locks.
- Shift all appropriate animals out onto their exhibits.
 - Radio another staff member advising them who/when animals are shifted into enclosure and yards.
- Update habitats, den fronts with green (empty) or red (occupied) magnets and move any cage identification magnets if needed.
- Mark down on dry erase boards in each area individual's locations and change cage cards to reflect what animal is in each habitat and/or den area.

Cleaning Night Houses

- Check weekly task lists for full cleans.
- All cleaning may be completed in any order most convenient to the animal care staff for that given day.
- Follow any LOTO SOP for entering and cleaning indoor dens.
 - Recalls for feeding.
 - Remove feces and wet/dirty straw.
 - If not already done and checked off on the task sheet, clean water bowls with soap and water daily. Refill water. Water bowls should be disinfected at minimum once per week.
 - Hose daily and disinfect at minimum once a week.
 - Replace hay beds as needed; hay beds should be replaced any time they are soiled.
 - Complete miscellaneous daily, weekly, and monthly general cleaning tasks.

General Tasks

- Upon completion of morning cleaning of animal areas, any miscellaneous tasks may be completed.
 - Examples: clean bathrooms, staff areas and hallways, kitchen area and sinks, refrigerators, graphics, stock shavings/grain/hay/meat/etc., take out trash, projects, etc.
- Make sure cameras are on and working, if not, please address the problem with the Head of the Animal Husbandry.
- Training
- Enrichment (put out new, clean old/used enrichment and store by the end of the day)
- Records should be filled out daily and throughout the day, so information is not lost.
- Fill out reports.
- Meetings
- Prepare diets (defrost two days in advance as per feeding charts). Record cold storage temperatures daily. Follow proper food handling and thawing procedures.

Closing Duties

- Finish setting up dens with evening diets, enrichment, bedding for all animals (if needed).
- Double check that all primary containment doors are locked and secured.
- Re-call animals off exhibits and feed PM diets.
 - If an animal does not shift in, notify the manager.
 - Double check that all animals are accounted for and secured in their yards (if allowed access to them to each other) and indoor dens.
- Turn off water features (if needed).
- Mark down on dry erase boards in each area individual's locations and change cage cards to reflect what animal is in each den.
- Update exhibits, yards, and den fronts with green (empty) or red (occupied) magnets.
- Finish any records.
- Wash dishes and put away clean dishes.
- Final life check should be done within the last hour of your shift.
- Verify cameras are still working and on.
- Double check locks.
- Turn off lights.
- Email daily report to Colossal husbandry team, managers and veterinarians.
- Put the radio and bear spray away.
- Clock out.

Record Requirements

Records are an important part of animal husbandry and veterinary care. Records should be kept on every Colossal animal for the entirety of their life cycle, including but not limited to: creation, use, preservation, and disposition of. All government permits, licenses, import, export, and transfer permits should be included in the animals' records.

Colossal's animals' records will be kept in the animal care information management system, which will include but is not limited to, daily husbandry records, individual records on behavior, enrichment, training, medical procedures, and all veterinary care information. Records should include, when necessary, the reasoning or rationale for decisions to help support transparency and allow for animal caretakers to understand the individual animals' history to ensure the animals are receiving the best welfare possible.

Vital records are to be archived and kept permanently. Vital records are any record or part of a record needed to reestablish care in an emergency. Records for deceased and disposition animals are vital to being able to track lineages, genetics of their descendants, and allow for knowledge of veterinary and husbandry practices to continue to evolve into best practices for the species and individuals in Colossal's care.

Veterinarians and veterinary technicians should record and enter all medical procedures, including but not limited to, visual examinations, routine examinations, and emergency procedures. All veterinary records will be entered into the animal care information management system by the veterinary team, or if a paper document is used, the paper will be scanned and included into the record and the veterinary team can attach it to the animal's medical record.

Animal care technicians, managers, curators, or any other animal care staff will use the animal care information management system to enter in and log daily husbandry for each animal and the animals' living spaces, behavior observations, and all enrichment and training information, including but not limited to, cleaning schedules, calendars, training sessions, shaping plans, approved enrichment, evaluation of enrichment, maintenance to habitats, barns, and yards, and all welfare concerns and evaluations.

Any communication regarding the individual animals and/or the group of animals will also be entered into The IMS. Animal care staff, along with the veterinary team, will be responsible for recording any life events, pregnancy, and results of pregnancies including termination of and reasons. Any offspring will be recorded regardless of its viability and lifespan, including stillbirths, abortions, premature birth, and failure to thrive situations. Information should be recorded into The IMS within two weeks of the event occurring. Photos and videos should be added, when possible, to help in capturing the information accurately.

All Colossal animals will receive an individual animal ID, and the animal or species will be included in the <u>Preventative Medicine Program</u> (domestic dogs will have a written veterinary care plan in lieu of, or in addition to, the Preventative Medicine SOP).

Animal records will include:

- Identifying marks, including scars, color patterns
- Tattoos
- Tags and location
- Breed/species
- Sex
- Age or estimate if unknown
- Name if they have one
- If an animal is microchipped, the number and location of the chip
- A picture of the animal, preferably a head on picture along with one or both side views
- Acquisition
- Breeding events
- Offspring
- Transfers
- Dispositions
- Reintroductions

Medical records will include:

- Date when problem was identified
- Description of the problem
- Examination findings
- Test results
- Treatment procedures performed and the results, if applicable
- Names of all vaccines administered and date of administration (if possible, site of administration to be able to track reactions)
- Names of and treatment administered and the date of administration (if possible, site of administration to be able to track reactions)
- Date and findings of any results of all:
 - Screenings
 - o Routines or other recommended tests
 - **o** Examinations
- Euthanasia records
- Necropsy procedure findings and results
- Histology and pathology results

Domestic dogs must have a written exercise plan, attending veterinarian approved exercise exemption, and a health certificate for transport.

Based on USDA recommendations, records should include the following:

- Attending veterinarian approval for non-commercial diets
- Enrichment logs

- Enclosure, habitat, and barn/holding areas cleaning and sanitation logs and/or records
- Enclosure, habitat, and barn/holding area maintenance logs and/or records
- Standard operating procedures, policy and/or protocols

TheHead of Animal Husbandry and Husbandry Manager will be responsible for going over daily animal care technicians' reports. Any items of concern will be addressed in the weekly meetings and a course of action will be planned if determined necessary.

All parts of Colossal animals, excluding feces and urine discarded in routine husbandry, should be documented and recorded as to why and how it was collected, where it ends up, and how it gets there so any parts of a Colossal animal are tracked and recorded into The IMS or the appropriate and agreed upon alternative for laboratory studies and work. This shall include, but is not limited to, animal paw/nose prints, hair samples, lost teeth, toenails, and blood and tissue samples. Any party interested in obtaining any part of a Colossal animal must receive written permission from the Head of Animal Husbandry. The information about who received it and the purpose for the request should be recorded into the IMS.

All veterinary procedures need to be recorded, and anything that can provide documentation of the welfare status of the animal, including but not limited to, changes in behavior, appearance, group behavior, scars, life events, cage moves, life/death events, and any information as to why decisions are made and how they were made objectively.

For example, we moved Dire Wolf Q to Habitat 101 because Dire Wolf A continued to be aggressive toward Dire Wolf Q, including biting at Dire Wolf Q, pulling out hair, and dominating food resources. Dire Wolf A is only aggressive towards Dire Wolf Q and is the dominant member of the group, so it was determined that moving Dire Wolf Q was better for the group than removing Dire Wolf A. Over the past few months we have added additional dens, feeding areas, and tried separation feeding individuals. None of the tried options seemed to help the situation.

the animal care information management system allows us to record the daily husbandry and enrichment aspects, and to assess their effectiveness, with each individual and group. Monthly and yearly reports should be run to monitor trends with individuals and groups. Any trends affecting an animal's welfare status should be addressed as soon as possible within the outlined communication methods and recorded in the animal's records.

Tips for Writing Records

Records should be complete enough so that if someone reads about the animal or group they would know the how, what, when, and why even if they haven't worked with the animal. The records should provide context for the person working with the animal or looking at the records one week, one month, one year, and/or 10 years from now. If you can't understand what you did with an animal by reading the record one month from now, most likely no one else will either.

When writing records:

- Re-read what you write.
- Edit your writing and remove any subject or emotions you may be inadvertently adding. Think about if you are focusing on the facts and animal welfare objectively or if you are allowing your personal feelings to color your record of the event. If the notes are not helping us make informed decisions regarding the animals in our care, the person writing the records should reword the information and

make sure the intent is having the impact of being objective and communicating all necessary information.

• Don't delete or not record events because it might require more time or typing to get the information recorded. Ask supervisors or fellow animal care technicians to help you find a different way to phrase the situation or shine some perspective on the event or animals. They may also be able to add some more information to offer clarity to the records.

Diet and Nutrition

The Colossal dire wolves' diet will be modeled after the grey wolves' diet. According to the AZA Mexican Gray Wolf Husbandry Manual, Mexican grey wolves should receive a fresh, high-quality diet. If changes need to occur in the diet, the changes should be gradual and recorded in the animals' daily records.

Diets should be evaluated by the Head of Animal Husbandry, supervisors, veterinarians and if needed, by a nutritionist anytime the BCS is out of the 4-6 range, the individual reaches a new life milestone, has a medical issue, or if their behavior changes. Diets may be reviewed at any time based on an animal's daily records and in accordance with the <u>General Communication Guidelines</u> during weekly meetings and site visits.

For Colossal dire wolves in managed care, nutritional requirements can be reached by feeding a good quality dry dog food such as Purina Pro Plan and Hill's Science Diets that are meat based, not cereal based. Food should be between 22-28% protein, 8-18% fat, and 2-4% fiber. Good quality protein should make up 20-25% of the dire wolf 's diet and fat levels should be at least 5% dry volume. Mineral and vitamin toxicity and deficiency are rare in dire wolves fed high quality diets. Most issues were found to be in dire wolves fed a diet of mainly organ meat or whole muscle diets. Feeding prepared meats and carcasses supplementally, in limited quantities, can be used as treats or for medicating and training routines.

Fast Days

Dire wolves can be given a fast day during the week, if there is not sustained extreme cold weather. Dire wolves should never receive fast days while there are pups present. Fast days are days in which beef/horse or other ungulate shank/knuckle bones are provided instead of the regular meat/dry diet. These provide some enrichment and good dental health to help strengthen cranial muscles and bones. Softer bones such as horse tails, chicken necks, and small animal carcasses such as rats, rabbits, chicken, and fetal calves can be fed periodically and can help provide calcium to growing, lactating, and nursing females. Whole prey and bone items should be sourced from a pathogen-free supplier and amounts fed should be monitored to prevent obesity or nutritional deficiencies.

Diet for Adults

Diet can be fed once a day. Pairs and groups do not need to be separated if the feed stations are set up to allow all members of the pack to eat their portion without a dominant member monopolizing feed and feed stations. If fed in a group, animal care takers should monitor behavior and consumption of all animals closely. Diet can be split up and fed multiple times a day to help with managing the dire wolves in regard to shifting into holding spaces, crate training, and for enrichment. If dire wolves are fed in the evening, the feeding should be given at least 40 minutes before animal care technicians leave for the day, so uneaten food can be removed and consumption amounts can be recorded if diet needs to be pulled to reduce the presence of pest species in the area such as raccoons, foxes, skunks and feral cats. Care should be used when applying the practice of removing diets at the end of the workday as dire wolves are very active at night and may consume most of their diet overnight. Removing the diet could unintentionally reduce the amount of food the animal consumes, by reducing the time the animal can eat.

More shy dire wolves may only consume their diet at night when there's less human activity. Feeding overnight may also need to be employed to help reduce competition from avian scavengers that are more active and can eat most of their diet; dire wolves rarely tend to run off avian species from their diets. Regardless of which feeding routine works the best, camera and video can assist in recording and monitoring how much diet each individual dire wolf consumes. Diets should be fed using a feed chute or placed into unoccupied secure stalls. Once staff is securely out of the stall, the dire wolf can be shifted back into the stall and secured while eating. All stalls, holdings, and outdoor habitats should have automatically refilling stainless steel water bowls. These are preferred, but dump and fill water bowls can be used so animals have water at all times. Large water troughs can be used if dire wolves don't have access to hoses or other chewable parts. Large troughs should not be used when pups are present. Need to add data about whole prey and carcass feedings

Diet for Pups

Once pups are being fed regurgitated food, a combination of dry dog food and prepared meat will help the regurgitated food have substance. Regurgitated dry food turns to mush and it will be harder for the pups to learn to chew and carry food. Typically, all members of the pack will feed the pups, so the pack's diet should be increased to accommodate the sharing of their portions when young pups are present. Growth requirements for pups are twice the maintenance diet requirement of an adult dire wolf. The adult pack members weight and Body Condition Scores should be closely monitored while pups are present to ensure all members and young are in good health and condition.

Food Storage

Dry diet should be purchased no more than three months in advance and stored in a cool, dry, pest-proof room or building with the label and expiration date facing out.

Prepared meats should be stored and thawed according to the guidelines from the AZA Nutrition Advisory Group.

Disinfection and Cleaning

Disinfectants are an essential part of disease prevention and are used to eliminate pathogens. Most disinfectants require direct contact with pathogens and areas to work effectively. Regardless of which chemicals are used, keep Material Safety Data Sheets (MSDS) or Safety Data Sheets (SDS) near where the chemicals are used, so they can be read before use and if inappropriate contact with chemicals occurs.

- Cleaning and disinfection are a two-step process. Cleaning should occur first and involves removing all the organic matter, such as dirt and debris, food, urine, and feces with detergent such as Dawn soap prior to the application of the disinfectant.
- After cleaning with a detergent, the area must be rinsed to remove the debris and the detergent prior to disinfection. Any organic matter left will inactivate disinfectants.
- Immediate removal of waste prior to disinfection is imperative so that re-introduction of pathogens and contaminated debris does not occur.
- Manufacturer's directions for dilution and usage will be followed exactly. This includes necessary protective gear that must be worn during usage of disinfectants. Some pathogens may require additional instructions, which will be determined by the veterinarian.
- The disinfectant must remain in contact with the surface for at least 10 minutes.
- Schedules will be established for regular cleaning and disinfecting in each area. Schedules will be updated and maintained in the animal care information management system.
- Footbaths must be used at entrances to Colossal animal buildings or holding areas. A boot brush, boot mat, or hose should be used prior to the footbath to remove dirt/debris which can inactivate the disinfectants.
- Footbaths should contain appropriately diluted disinfectants and must be changed regularly, regardless of disinfectant used. They must be changed at a minimum of once a day but may need to be changed after every use. If the footbath is 10% organic material (dirt, feces, etc.), the disinfectant will inactivate, and no longer be effective. If bleach is being used as a footbath, it must be at least 15% bleach solution.
- Bedding should be clean, dry, free from urine and feces, insect-free, smell of wolf but not mold or other foul smelling odor and changed as needed. All bedding should be checked thoroughly daily, and animal care technicians should record when it is changed in the animal care information management system.
- Staff areas including cabinets, shelves, closets, and counters should be free of clutter that may provide shelter for rodents and insects.
- Animal contact with potentially harmful chemical agents should be prevented in all cases.
- All waste/debris and fecal containers should be cleaned and disinfected at a minimum once a week. All waste and fecal matter should be promptly and appropriately disposed of. Special considerations include:
 - Enclosures without substrates should be cleaned and disinfected daily or on a regularity agreed upon by the veterinarian and Head of Animal Husbandry.
 - Enclosures with hay substrates should have all feces, urine, and the top layer of hay substrate removed daily. Spot cleaning and disinfecting should occur daily. Substrate should be completely removed, and the enclosure disinfected as directed at least 1-2 times weekly.
 - Enclosures with natural substrate floors should have feces removed daily. Spot cleaning and disinfecting should occur daily. Any urine puddles should be bailed out or filled in with additional substrate. The substrate should be completely removed 1-2 times per year.
- Food containers should be cleaned and disinfected daily. Thorough rinsing is essential. Large food troughs and water troughs should be cleaned daily and disinfected at least once weekly.
- Food preparation areas, barn hallways, and work areas must be cleaned and disinfected daily.
- Non-animal areas and unused holding areas must be checked daily and cleaned when needed and before animals are moved into an unused space and after they are moved out of the space.
- Appropriate personal protective equipment (PPE) must be worn while cleaning, disinfecting, and hosing to

reduce the animal care technician's exposure to fecal material, infectious agents, and chemicals.

- Disposal of any expired cleaning solutions should be done as directed by the manufacture of the product:
 - 1) Rescue Ready-to-Use can be poured down the drain according to the manufacturer.
 - 2) Rescue Concentrate should be diluted and poured down the drain according to the manufacturer.

Below are examples of disinfectants that may be found at Colossal Biosciences.

Disinfectant	Туре	Spectrum	Dilution	Cont act time	Effect	Corrosive
F10 SCXD	Quaternary ammonia and biguanide	Bacteria, mycobacteri a, viruse s, fungi	1:200 5 ml: 995 ml	10 min	Cleans, disinfects	YES1
Oasis 146	Quaterna ry ammonia	Bacteri a, viruses , fungi	1:128 1 oz: 1 gal	10 min	Disinfect s	YES ¹
Roccal	Quaterna ry ammonia	Bacteri a, viruses , fungi	1:256 ¹ ⁄ ₂ oz: 1 gal	10 min	Cleans, disinfects	YES ¹
Bleach (sodium hypochlorit e)	Halogen 5.25-6.15 %	Bacteri a, viruses , fungi, protoz oa	1:32 4 oz: 1 gal	10 min	Disinfect s	YES ¹
Calcium hypochlorite	Halogen	Bacteri a, viruses , fungi, protoz oa	1 g: 15 gal	10 min	Disinfect s	YES ¹
Chlorhexidine (Nolvasan)	Biguanide	Bacteri a, fungi, some viruses	1:128 1 oz: 1 gal	10 min	Cleans, disinfects	No
Stable Environment	Enzymatic	None – cleans only	Variable	5 min	Cleans	YES ¹
Accelerated hydroge n peroxid	Oxidative	Bacteria, viruse s, fungi	0.1-0.3%	10 min	Cleans, disinfects	NO
Virkon-S	Oxidative	Bacteria, viruses, fungi	10 g: 1 liter	10 min	Disinfect s	YES1
Buckeye Eco Neutral Disinfectant	Germicidal detergent	Bacteri a, viruses , fungi	1:256 ½ oz: 1 gal	10 min	Cleans, disinfects	YES1

Simple Green All- Purpose Cleaner	Non-ionic surfactant	Bacteri a, viruses	1:1	2 min	Cleans, disinfects , degreaser , deodoriz es	NO
Rescue Ready-to- Use	Hydrogen peroxide oxidative	Bacteria, viruses, fungi, protozoa, mold, mildew, allergens	None	1 min	Disinfect s, deodoriz es	NO

¹ Causes irreversible eye damage or skin burns. Harmful if swallowed or absorbed through skin. Do not get in eyes, on skin or on clothing. Avoid breathing spray mist. Wear protective eyewear, protective clothing, and rubber gloves when handling.

Behavioral Husbandry Policies and Procedures

Behavioral husbandry is a component of daily animal care and welfare and influences the quality of an animal's life in its habitat.. Behavioral husbandry focuses on how animals interact with their physical and social environments. The goal of behavioral husbandry is to support holistic behavior management utilizing training, complex environments, and enrichment. To fulfill the goal, we must understand the natural history of the species, the individual's history, and behavioral science. Colossal will provide Colossal's animals with an environment that allows animals to be well adjusted, demonstrate natural behaviors, and successfully cope with and adapt to changes in their environment that mimic or go above their wild counterparts. The program will be continually evaluated to ensure its success.

Colossal uses behavior analysis to study the individual's behavior in relation to their environment using sight, sounds, smells, touch, etc. Using what we learn about the individual's relationship and environment to change the individual's behavior is called applied behavior analysis.

Behavioral husbandry is considered part of the animal care technician's daily routine. Colossal's Behavioral Husbandry Program incorporates the safety of animals and staff and is based on programs from:

- AZA facilities
- The Association for Behavioral Analysis International
- Disney's Animals, Science and Environment Animal Enrichment Program
- Animal Training Fundamentals with Barbara Heidenreich
- Dr. Isabella Clegg, Founder and Director of Animal Welfare Expertise
 - Welfare Assessment Template is located in the animal care information management system.

Colossal's Behavioral Husbandry Team includes the following:

• The Head of Animal Husbandry is responsible for the oversight of the behavioral husbandry program including enrichment and training. The Head of Animal Husbandry will approve all enrichment and training programs for Colossal animals and ensure that all facilities follow the SOPs, communications, budgets, and support facilities, and liaison with the veterinarians.

- **The Manager(s) of Animal Husbandry** is responsible for developing, guiding, and continuity for animal training and enrichment programs at all facilities. The Manager of Animal Husbandry will support the Head of Animal Husbandry and staff, review training and enrichment submissions, and assist facilities with documentation.
- The Director of Animal Health will assist in evaluating and approving the medical aspects of behavioral husbandry programs including enrichment and shaping plans to ensure animal safety and that the highest standards of medical care are met. The Head of Animal Husbandry will be responsible for consulting with the veterinarian if needed.
- A nutritionist will assist in evaluating and approving the nutritional aspects of behavioral husbandry programs including enrichment and shaping plans to ensure animal safety and that the highest standards of nutrition are met. Head of Animal Husbandry will be responsible for selecting and consulting the nutritionist if needed.
- Animal care technicians and supervisors will work together to identify and create goals, shaping plans, and enrichment. They are the eyes and ears for the management team and key to the communication process. They are responsible to ensure that animals in their care receive the best standard of care and welfare possible.

Behavioral husbandry will be reviewed with the Colossal Husbandry Manager(s) and Head of Animal Husbandry during routine weekly meetings and bi-annual site inspections.

Animal care technicians will be able to submit new enrichment and training ideas for review and approval to the Head of Animal Husbandry and Manager of Animal Husbandry. All existing training and enrichment will be reviewed yearly, or as needed, to make sure they are effective for the individual animals and/or the group of animals. No training/shaping plan or enrichment device or plan should be utilized on/with an animal without Colossal approval.

Animal Training

Colossal will identify specific behaviors to facilitate husbandry and veterinary procedures through the animal's voluntary participation whenever possible and feasible. The program will be goal focused and utilize shaping plans developed utilizing the S.P.I.D.E.R. model:

Setting goals Planning Implementation Documentation Evaluation Re-adjustment

Training and shaping plans should utilize the natural history of the species and the individual animal's history and personality. Shaping plans will be evaluated and adjusted throughout the process based on the individual animals, animal care technicians, and facilities' progress in the shaping plan. Colossal recognizes that each individual animal, species, and animal care technician team will have different successes, learning curves, and challenges. Colossal will celebrate, support, and hold accountable the facilities working through the shaping plans utilizing the communication standards outlined in the S.P.I.D.E.R. method. Operant conditioning can

provide animals with mental stimulation that mimics natural behaviors and challenges their wild counterpart's encounter. Thus, training is a component of behavioral enrichment as well as an input for animal welfare.

Enrichment Programs

Colossal enrichment programs should start with creating complex environments that encourage and allow for species specific natural behaviors. In addition to their environment, animals will be provided with activities throughout their day to allow them to utilize their species appropriate behaviors, while giving them choice and control in various aspects of their lives. Enrichment programs will be developed utilizing the S.P.I.D.E.R. model, species natural history, individual behaviors, and the identified behavioral goals. The program will be approved and evaluated to make sure it increases desired natural behaviors or reduces or eliminates inappropriate behaviors. This will be done utilizing the communication standards outlined in the <u>General Communication Guidelines</u>. Enrichment programs will be reviewed by the of Animal Husbandry for safety before being given to any Colossal animal.

The enrichment program should:

- Be behavior based
- Enhance animal welfare
- Promote species appropriate behaviors
- Address and modify behavioral needs and issues

Behavior/Enrichment Categories

- Resource Acquisition
 - o Locating, obtaining, consuming food/water in a natural method
 - Examples: foraging, scent trails, climbing, ice blocks, puzzle feeders, contra feeding
- Rest
 - o Encouraging natural inaction or development of a resting area
 - Examples: basking/sunning, utilizing temperatures and heating devices or natural light, nest making, den digging, perching
- Avoidance
 - o Offensive/defensive behavior to avoid predators or aggressive conspecifics
 - Examples: hiding, head butting, charging, mock charging, fleeing from enrichment, being on alert
- Territorial
 - o Protecting a resource such as food, water, conspecifics, or an area of habitat
 - Examples: food caching, vocalizations, scent marking, defining territory by scent or trail running
- Social
 - o Creating opportunities for social interaction
 - Examples: grooming, playing, babysitting, play fighting, conspecific and allospecific interactions
- Communication/Senses
 - o Signaling between two or more animals through vocalizations, non-verbal behavior including scent marking, body language
 - Examples: introducing mirrors, smells, sounds and allowing group hunting

- Investigative Play
 - o Encouraging species specific behaviors such as hunting, defense, stalking
 - Examples: provide stimuli to encourage exploration and play behaviors
- Environmental Change
 - o Controlled change of environment to mimic season, migration, rainy season
 - Examples: pool changes, new perching, new logs, rotating habitats, dry season, sprinklers
- Self-maintenance
 - o Encouraging self-care behaviors for the animal such as shedding coat, coat grooming, wallowing
 - Examples: brushes, dust/sand, mud wallows, soft animal scratching pads, logs, boulders, insect repellant fire hose flaps
- Parental/Courtship
 - o Behavior to attract a mate or to help ensure offspring survival
 - Examples: nest building materials, egg sitting, allowing mate choice, corearing, mirror or sound to stimulate large herd/flock sizes

S.P.I.D.E.R. for General Behavioral Husbandry

Utilizing the industry standard of S.P.I.D.E.R. for both enrichment and training requires the use of goal-setting questions to understand the species' natural history and the individual animals' history. Colossal's goal is that each enrichment/training plan has a natural/individual history form completed and approved by the Colossal Head of Animal Husbandry. Our behavioral husbandry program is goal-based vs. item based. We will use items to increase desired behaviors and decrease maladaptive behaviors.

Facilities will use the S.P.I.D.E.R. model to maintain a consistent process.

S.P.I.D.E.R. for Behavioral Enrichment

S. Setting Goals

- Learning the natural and individual history of the species
- Clearly identifying the behaviors we want to develop or to discourage through enrichment and/or training

We will adjust the goals throughout the animal's life depending on their biological, social, medical, and cognitive needs. When setting goals, staff should consider the frequency in which enrichment will be utilized, including seasonal, daily, weekly, monthly, and yearly. If there is limited recorded information on the species in question, please see the Head of Animal Husbandry for assistance.

Natural/individual history forms can be found here.

P. Planning

- Which behaviors are to be encouraged/discouraged?
- What resources are needed to create the enrichment?
- Where is the enrichment going to be utilized?
- What are the safety concerns?

Planning involves the creation of enrichment to allow animals to have choice and control within their environment. The planning process should help animal care staff think through the idea and how it will increase desired behaviors and decrease undesired behaviors. Animal care technicians, supervisors, curators, and owners along with the Colossal husbandry team will be involved in brainstorming ideas for enrichment and training plans. All staff members are welcome to provide ideas and suggestions following the S.P.I.D.E.R. model. Undesired behaviors may require further observation, and an action plan may be utilized to decrease the occurrence or eliminate the behavior.

No enrichment item/plan/training plan should be started or given to an animal without prior approval from Head of Animal Husbandry. The enrichment plan/training plan must have the appropriate approval form filled out; the <u>Natural History/Individual History Form</u> should be included when submitted to the Head of Animal Husbandry. <u>See Enrichment Approval Form</u>.

I. Implementation

- Implementation should be planned in advance.
- Implementation of the plan includes scheduling enrichment in the calendar, ensuring the enrichment is available, cleaned, and in good condition.
- All items and plans must be approved by the Head of Animal Husbandry in advance through the <u>Enrichment Approval Form</u>.
- To be most effective, the enrichment should be changed out often and varied. The purpose of enrichment is to increase desired behaviors and reduce or eliminate maladaptive behaviors. It is intended to increase the welfare of the animals, not to check a box, or meet the personal needs of the animal care technician.
- Novel enrichment should be integrated in all categories.
- If multiple animals are in the group, there must be enough items for each individual animal of the group.
- Enrichment must be recorded in the daily records in the animal care information management system.
- Evaluate, reassess, and adjust as data suggests and anytime a change is made to the animals' social, medical, or welfare status.
- Colossal will utilize veterinarians and nutritionists to approve/adjust and monitor food enrichment and training rewards. The Head of Animal Husbandry will be responsible for working with them and determining quantities and frequency.

D. Documentation

All animal care technicians are responsible for documenting enrichment and training. They will document the enrichment/training in the animal care information management system on the day it is given. This should include how the animal interacts/participates with the enrichment/training program. Enrichment schedules, enrichment plans, training plans, and shaping plans will be uploaded and tracked in the animal care information management system. Videos and photos will be taken and uploaded into the software. Trends can be identified and monitored to make sure they are effective and meet goals. Animal care technicians may not be able to observe animals interacting with enrichment but may be able to deduce that the animal utilized the enrichment. They should include if they directly or indirectly observed the enrichment being utilized.

E. Evaluation

Evaluation will be a multifaceted approach including observations, videos, photos, forms, meetings, and the animal care information management system. This approach will evaluate if the enrichment/training plan achieves the desired behavioral goal. Facilities and Colossal will evaluate the trends and patterns and how the animals utilize the enrichment to determine the effectiveness and whether to continue utilizing or discontinue the enrichment. Every time enrichment or training plans are utilized, the effectiveness should be notated and management can evaluate the notations with during weekly meetings. Enrichment and training programs will also be evaluated during the Animal Welfare Assessments. Staff will use the <u>Enrichment Assessment Form</u> to evaluate the enrichment plan.

R. Re-adjustment

Enrichment and training plans can be readjusted at any time before, during, or after the S.P.I.D.E.R. process. Adjustments will be noted in the enrichment and training plans. Adjustments may result in discontinuing of or modifying existing enrichment and training/shaping plans. Reassessments may also require new enrichment to be considered to ensure behavioral goals are being met.

Behavioral Training

Ken Remirez, author of "ANIMAL TRAINING: Successful Animal Management through Positive *Reinforcement,*" explains that training is teaching. Similarly, Colossal considers behavioral training as developing a method of communicating and teaching between two people, two animals, or a person and an animal. Reasons that behavioral training is part of behavioral husbandry are safety, decreased stress, proactive medical care, exercise and mental stimulation, and research. Colossal's goal is to improve animal welfare by integrating behavioral training into daily animal management. It is a component of daily husbandry.

Every direct and most indirect interactions between an animal care technician and the animals can be a form of training. Being mindful of this while working with and around the animals in our care will help to ensure that what we teach the animals encourages goal behaviors and not maladaptive behaviors. The problem solving associated with operant conditioning can help create the mental stimulation associated with the species' natural behaviors. Training engages animals on a cognitive level and can increase the animal's positive welfare. It can increase positive animal care technician interactions and assist with routine husbandry and medical behaviors by reducing stress to animals and staff and reducing time to accomplish these tasks.

Training is a component of enrichment. The S.P.I.D.E.R. model should be utilized when creating training/ shaping plans for all animals. All animal care technicians are expected to tell the truth in all documents, so goals can be reached and for the best welfare of the animal. Adjustments can't be made correctly without accurate information and documentation. <u>Karen Pryor's 10 Modern Principles of Shaping</u> are a great resource to use during the S.P.I.D.E.R. process.

Colossal expects every trainer to:

- 1. **Be honest and forthright.** If the members are not truthful, the team cannot be clear in the methods and goals, and this can lead to misunderstandings.
- 2. **Be consistent.** Each animal will have a set of animal care technicians that are considered primary trainers. Each individual behavior should ideally have one trainer. Colossal understands and supports that some species will require more than one primary trainer. Secondary trainers will serve to maintain the behavior(s) when the primary trainer(s) are not available. All members of the training team should be aware of the standards and levels of progress the animal and team are making.

- 3. **Be self-aware.** While being self-aware may seem like the easiest thing to do, it is often the most challenging to communicate. An effective team is essential to the success of the training program. If we allow individual egos, triggers, and insecurities to manifest and not be addressed proactively, the team dynamic will be affected, the success and progress of the animal decreases, and the animal can even regress. The ego can be defined as the creator of stories, excuses, or the "worry about how we are perceived by others." Being self-aware includes communicating and addressing these thoughts, concerns, and feelings in advance, and when they occur, helps to eliminate power plays with animals and team members.
- 4. **Beware of dogma**. If you are doing the same thing repeatedly and expecting a different result, you may walk away for a short time, not communicate effectively, and/or give up entirely. This will not only stall out or shut down a training program, but it can also stall out or stop your development as an animal trainer.

S.P.I.D.E.R. For Creating a Training Plan

S. Setting Goals

- Learning the natural and individual history of the species
- Clearly identifying the behaviors we want to develop or discourage through training and/or enrichment

P. Planning

- Planning involves the development of a training plan that clearly outlines the steps of how the behavior will be shaped. Most of the time, the training plan may look very different from the original plan that was laid out. During this portion, the schedule of reinforcement or rate at which reward is given to the animal and the amount of reinforcement items should be determined and outlined. Colossal encourages a 1:1 ratio of reinforcement (one behavior asked for (cued), behavior demonstrated, one reinforcer given), but recognizes that each behavior, team, species, and animal will need to be considered and the best choice for that situation should be used.
- When deciding the schedule of reinforcement, consider the duration of the session. How long does the animal need to demonstrate/present the behavior (touching a target or holding a leg bent for hoof trim)? Does the shaping plan allow the animal for choice and control? For example, does the animal have to be secured in a restraint device for a blood draw or can the animal line up along the fence for a successful blood draw? Will giving the animal the choice to leave make the animal more comfortable and thus increase the chances of a longer more successful blood draw?
 - **Continuous:** Animal continuously receives reinforcement while demonstrating the correct behavior and every time it demonstrates the behavior when the behavior is cued.
 - Fixed Interval: Reinforcement is given after a set amount of time.
 - o **Fixed Ratio:** Reinforcement is given after a set number of behaviors.
 - o Variable Interval: Reinforcement is given after a varying length of time.
 - o Variable Ratio: Reinforcement is given after a varying number of behaviors.

I. Implementation

• Implementation includes execution of the plan and shaping the animal's behavior toward the agreed upon and desired goal.

D. Documentation

• Documentation is crucial as it deals with how information is communicated to the team, how the training sessions are progressing, and what is occurring during the sessions. This allows us to document trends, behaviors, and animals' preferences. It helps to determine at what step the animals are progressing or struggling during the shaping of the desired behavior. Training progress, including videos of training sessions, will be documented in the animal care information management system. This also allows for animal care technicians, supervisors, and Colossal to support the sessions to identify inconsistencies.

E. Evaluation

• Evaluation allows the training team and trainer to look back at trends over time, including the development of the trainer.

R. Re-adjustment

• The team can meet and make changes to the training plan at any point during the S.P.I.D.E.R. process. These adjustments can be used to help the animal achieve the goal behavior and serve as a reference for the animal for future behaviors.

Determining Training Behaviors

The behaviors to be trained will be prioritized with the supervisors and Head of Animal Husbandry. The species, the individual's needs, the needs of the group, and habitat will determine which behaviors are to be trained, frequency of training, and available resources. Training will be done in a protected contact setting unless it can be determined and approved by the Head of Animal Husbandry that free contact is completely necessary to reach the goal behaviors, and it is safe for the staff and the animals.

As a reminder:

Free contact is defined as when humans and an animal share the same unrestricted space; there are no barriers between human and the animal.

Protected contact is defined as an animal and human not sharing unrestricted space by use of some type of protective barrier. The animal is not confined or forced to interact and can choose to leave the work area at will.

The requestor will have to outline in detail the rationale why free contact is vital to the success of the individual animal to reach the desired behavioral or medical goal. Colossal will consult with experts as they deem necessary if they feel it is required for the decision. Time and ease will not be considered necessary unless it is a quality of life concern. Free contact comes with significant risks to human safety and can cause animal safety concerns and animal escape concerns. Free contact may be utilized for special cases such as hand rearing and will be outlined in the species-specific hand rearing SOPs. Most nondomestic animals (usually everything except domestic dogs and cats) prefer protected contact as it gives them control of their environment and allows them more choice, often providing better welfare options for the animal.

Training New Behaviors

Training new behaviors must be approved by Head of Animal Husbandry before any training and/or shaping of the behavior begins. Staff will complete the <u>natural history/individual history form</u>, along with a completed shaping plan. These plans will be reviewed in the weekly meetings before being approved and can be revisited at any time during the training process. Training plans must be filled out completely and in detail. Colossal will evaluate the plan for timing/staff commitment and make suggestions/adjustments before approving or denying the training plan. The approved and denied documents will be uploaded into the animal care information management system. No training should occur before a plan is approved. All training sessions are to be documented, and the documentation is the responsibility of the animal care technician who leads the training session. The animal technician is responsible for updating the list of trained maintenance behaviors and providing a list of staff that have been signed off for training, including the specific behaviors and animals that individual staff are allowed to train. This information should be communicated to the Head of Animal Husbandry and Husbandry Manager.

Documenting Training

All training sessions, regardless of success, should be recorded in the daily report and the training sections of the animal care information management system. The Manager of Animal Husbandry will work with the team to build the shaping plan into templates to assist in consistent documentation of the training sessions. Colossal's animal care team will work together to evaluate the shaping plans in real time to ensure the animal reaches their behavioral goals. The trainers should write a monthly report summarizing how the training is progressing and any successes or challenges and submit it to Manager of Animal Husbandry. Honesty and transparency are expected and required in all documentation.

Training is one of many animal management tools and an important part of animal husbandry and welfare. All animal care technicians and management are involved in the training of the animal whether directly or indirectly. Training plans should be proactive, not reactive. Trainers should review past sessions to look for and identify patterns. Past sessions may help predict the animals' reactions to new behaviors.

Measuring Success

Success will be measured by how well the animal is trained to the level that other members of the team can also have the animal successfully demonstrate the behavior accurately and comfortably. The success should be the animals and the teams, not the primary trainers.

Training is a dynamic and continually improving field. Colossal recognizes that behavioral management is evolving as industry and science improves. Colossal's training philosophy is to utilize science, innovation, and best practices to provide the best welfare to the animals in our care at all our partner facilities. Behaviors to be learned must be effectively communicated to the animals in a consistent manner to make the learning process go as smoothly as possible for the animal. When deciding which behaviors to train, it should be considered if the animal is physically capable of presenting the behavior. We will make sure the animal is motivated enough to learn and execute the behavior using positive reinforcement. These reinforcers can be adjusted as needed and with approval from Colossal and the supervisor. When behaviors need to be modified, we will start by ensuring that all the animal's nutritional, physical, and medical needs are met. Once these needs are verified to be met, we will follow the **hierarchy of behavior modification** starting with the least intrusive to the most intrusive.

- Meeting Nutrition, Medical, and Physical Needs
 - Is the animal in pain, distress, or is the animal hungry?



Creating a Culture of Safety

Training animals can assist in making their care safer for the animals and staff. However, it can on occasion place the animal care technicians in close proximity to potentially dangerous animals or in potentially dangerous situations. Colossal strives to create a safe culture in all areas of animal care by encouraging day-to-day awareness, procedures, and staying vigilant against becoming complacent in daily repetitive routines, training sessions, and procedures.

Situational awareness is a person's accurate perception of the reality of their current situation.

- When working with animals, animal care technicians are expected to read all training and enrichment procedures and check for changes every session/day.
- During training sessions, animal care technicians should constantly be aware of grab/squish zones, their positioning in regard to the animal and structures, and the animals' capabilities. They should never underestimate an animal or overestimate their own ability to respond faster than the animal.
- Animal care technicians should always have an exit/escape strategy in case of an emergency.
- It is expected and required for animal care technicians to communicate if they feel unsafe, unfocused, distracted, or need to end a session. Each trainer is responsible for honestly communicating to the participants in the training session to ensure that all participants, (including the animal) are safe and the training session is positive and successful. Some behaviors, areas where training occurs, or animals may require a spotter to ensure training success and safety.
 - Example: a trainer working on a foot command for the dire wolf cannot see the precursor behaviors to a wolf spooking, so a spotter should be utilized to watch the whole wolf and not just its one foot. This may require three trainers: one to reward the wolf, one to work near the wolf's foot, and one to watch the whole wolf and both trainers. The spotter should be able to make sure the trainer working on the foot isn't in a dangerous position while watching the wolf for precursor behaviors to ensure a positive session.

For a productive session, always take these safety considerations into account:

• Prep the area. Make sure trip hazards are removed, that people have clear access to move to safety, and that the area is free from distractions. Additionally, make sure that the animal's space is ready and free from distraction for the animal.

- All items, props, reinforcers, and the bridge should be in place for the session.
- Feeding method, feeding items, and rate of reward are stated in the SOPs and are to be followed by everyone. All trainers must be aware of the SOPs and in understanding and agreement of what is being outlined and communicated before starting the session. Example: Is the count 1, 2, 3 or 1 Mississippi, 2 Mississippi, 3 Mississippi?
- Team agreements are critical. The specific need of the animal and the goal should be mutually understood, agreed upon, and updated regularly as the animal and team evolve. This should be discussed in training meetings as well.
- Maintain situational awareness of environment, time of day, and length of session. If needed, get a spotter or adjust to ensure a positive and safe session.
- Communicate comfort levels. Trainers are required to communicate their comfort level with the animal and the team before sessions begin. If a trainer needs to call a session they should communicate with management as to why they called the session and their concerns as soon as they can.
- Know your animals, recognize precursors, and be aware of inadvertently reinforcing unwanted behaviors, aggression, and stereotypical behaviors. Know your species and individual animal and any social implications that may occur for the animal before starting the session. This should be discussed during the planning portion of the S.P.I.D.E.R. process.
- All trainers should be fully present and focused on the session. During training sessions, phones should only be used for video purposes and should not be used by trainers working with the animal. The session can be recorded by setting the phone to video on a tripod or having another animal care technician record a video for the trainers. Radios should be on but turned down and non-emergency calls should be ignored and returned after the session.

All animal care technicians should become aware of the Basic Training Terminology utilized by AZA and American Association of Zoo Keepers (AAZK). It can be found on both the AZA and AAZK websites.

Trainers should remember to:

- Focus on the behavior goal.
- Be innovative.
- Be present and give 100% to each session.
- Treat each animal as an individual and avoid generalizing.
- Observe all aspects, and observe the animal before, during, and after the session.
- Recognize that challenges are opportunities to learn and progress.
- Mistakes are opportunities to start anew with more information.
- Leave your ego at home—individuals don't succeed unless the team succeeds.
- Take responsibility for your animal's behavior and your own. Don't blame others or make excuses.
- Labels are not solutions. Speak in observable behavior terms, look for understanding and solutions. For example, calling an animal stubborn isn't helpful. But if the animal refuses to enter the chute, we can start to identify the reason why the animal won't enter the chute.
- Earn trust with the animal and never take for granted the working relationship with the animal and fellow animal care technicians.
- Build and develop the secondary trainers to increase trust in the animal and the team. This ensures the animals' progress and welfare doesn't decrease during staff vacations and as changes occur at facilities.
- Animal training and team building are not linear—they are a living, fluid combination of science, trust, and art.
- A behavior is never fully trained, we are always adjusting. As animals learn, grow and mature, the trainers learn, grow, and mature.
- Take initiative in continuing to learn and grow as a trainer and team member.

- Share what you learn.
- Be open to learn from the animal and from your fellow trainers.

The forms on the following pages are to be utilized for the behavioral husbandry programs. Forms should be completed and emailed to the Head of Animal Husbandry and Manager of Animal Husbandry. Forms will be reviewed and the senders notified of the outcome. Finalized forms should be uploaded into the animal's file in our information management system.

Natural History Form 2025 .pdf

Canid Individual History Jan 2025.pdf

Current Habitat Form Jan 2025.pdf

Enrichment Approval Jan 2025.pdf

Enrichment Assessment Form Jan



Natural History

Facility: Date: Species: Individual ID: Animal Name: Submitted By:

1. Describe the species' wild habitat. E.g., desert, tropical rainforest, cover, moisture, concealment/camouflage options, temperature ranges, barriers from conspecifics. (If specific information on a particular species is unknown, provide information on closely related species/genus/family.)

2. Describe the species temperature/humidity range in the wild. How do they respond in the wild to the changes in temperature, weather, and seasons?

3. What are the animal's self-maintenance behaviors? E.g., preening, grooming, dustbathing, wallowing, sunning. Do they have a seasonal molt or shed? If so, when does this occur?

4. When is the species most active in the wild? Is it diurnal, nocturnal, crepuscular? Why? (Food availability, predators?) Does this change with seasons or location, E.g., urban vs. country?

5. Is the species in the wild arboreal, terrestrial, aquatic? Or does it inhabit different environments at different times of its life, day, seasons?

6. Describe the animals' main threats in the wild. What is the species afraid of (humans, conspecifics)? What are the different predators it encounters in the wild?

7. Describe the antipredator behaviors for this species. E.g., killdeer with broken wing display. Where and how do the species take refuge from fearful situations in the wild? What does this species' fear behaviors look like?

8. What is the social structure of this species in the wild? What is the average/typical group size? E.g., solitary, colonial, harems, pair.

9. What is the average distance between individual members of a social group? What is the average difference between neighboring conspecific social groups?

10. What are the primary sensory modalities for communicating with conspecifics, detecting predators, finding mates, and food? E.g., sight, smell, hearing, touch.

11. Describe the primary social behaviors of this species. E.g., aggression, play, courtship.

12. How does the social structure change seasonally, throughout species life? E.g., juvenile vs. adult, bachelor groups, plentiful food vs. limited food resources.

13. How does this species defend territories? Does it maintain a home range? What is the size of the home range? Does the species migrate? If so, when?

14. How does the animal advertise/mark its home range/territory? E.g., scent marking, vocalizations.

15. How does this species attract a mate in the wild? Do both males and females display? Is it the same display?

16. Where and how does the species raise its young (nest/den, location types, number)? What materials does it use to build nests or dens? Are both sexes involved? Is the entire social group involved? Who is involved in rearing the young? Does that look the same for all individuals rearing the young?

17. Are young precocial or altricial? How are the young fed? Who is involved in rearing the young? Does that look the same for all individuals rearing the young?

18. What is the species diet in the wild? E.g., carnivore, omnivore, herbivore, insectivore, fructivore. Does it change seasonally, by age, by location?

19. What variety of food does the species eat in the wild? E.g., different grass types, types of trees, does it only eat leaves or bark, what species of animal does it eat? Does this change seasonally, by age, by location?

20. How does the animal locomote through its habitat? E.g., swing through trees, swim, walk.

21. Where and how does the animal sleep or rest? For how long? Does it change seasonally, by age, by location?

22. Any other considerations for this species?

Individual History

Facility: Date: Species: Individual ID: Name: Current Habitat: Other Animals in Habitat: Other Species in Habitat: Submitted By:

1. Does this individual animal have any medical problems? If so, please describe. E.g., arthritic, obese, diabetic, pinioned.

2. Does this individual animal have any behavioral problems? If so, please describe. E.g., aggressive to humans, imprinted on humans, feather plucking, stereotypies.

3. Does this individual animal have any natural behaviors that it is not doing? If so, please describe.

4. Any other considerations for this individual?

Current Habitat

Facility: Date: Species: Individual ID: Name: Current Habitat: Other Animals in Habitat: (include population numbers, e.g., 1.5 wolves)

Other Species in Habitat: (include populations)

Submitted By:

1. Describe the size and shape of the animal's enclosure. Are there tight corners? What is the size and shape of the holding/barn/stalls?

2. What are the containment barriers?

3. Can the animal use all components of its exhibit? Can it hide? Are there ways for it to be out of sight of cage mates?

4. How functional/dynamic is the current habitat? Does the habitat encourage/allow the animals to exhibit natural behaviors? How does the animal interact with the exhibit elements? Include all behaviors both adaptive and maladaptive.

5. Where and how is the animal's food provided? Does the animal show a preference?

6. Does the physical environment contain elements of novelty? Can the furniture be changed? Can weather changes be simulated?

7. What are the animals' opportunities to feed, forage, breed, and socialize in species appropriate ways?

8. Can/should/does the animal interact with other species in the habitat?

9. Can the animal exhibit normal patterns of behavior? Does the physical environment allow for this? E.g., Can the duck float and swim in a pool deep enough that its feet don't touch the bottom?

10. Can the animal make choices about where and how it spends its time? How does the animal demonstrate this?

11. Can the animal have control over its acquisition of food? Access to hiding places?

12. Does the animal have access to areas allowing it protection from the elements?

13. What are the hazards in the habitat?

14. Given these considerations (the natural history, individual history, current enclosure) what behaviors are being encouraged? Discouraged?

Enrichment Approval Form

Facility: Date: Species: Individual ID: Name: Submitted By:

Describe proposed enrichment. Include photos and suggested frequency.

Describe the goal of the behavior the enrichment will affect.

Where will the item be given: on habitat or in holding?

How will the item be put into the area where it will be used? How can the item be removed from the area? How can it be removed in case of an emergency or unforeseen issue?

Describe safety concerns and ways to mitigate those concerns.

Risk Assessment

Does the item pose any danger to the animal? If so, please describe.

Does the item pose any danger to enclosure mates? If so, please describe.

Can it fall on any animal in the habitat? If so, please describe.

Can it break any part of the habitat? If so, please describe.

Can it break or cross containment barriers? If so, please describe.

Can the animal throw this at people or other animals? If so, please describe.

Will the item cause displacement or fighting amongst animals on exhibit? Would more items mitigate this risk? If so, please describe.

If the enrichment is food, is it a reasonable amount? Is it safe for the animal? If so, please describe.

Can the animal choke on the enrichment? Can an exhibit mate choke on the enrichment? If so, please describe.

Will the item traumatize the animal or exhibit mates? If so, please describe.

Has the item already been approved or denied? If so, please describe.

Can the item be used for any animal in the habitat to escape? If so, please describe.

Do the benefits outweigh the risks or do the risk outweigh the benefits? If so, please describe.

Acquisition of Item

Where can items be purchased? (include model/part number, hyperlink, number of items needed)

Who will construct the enrichment item if applicable?

Please share the website or email of the contact for purchase.

Describe how the effectiveness of the enrichment plan will be evaluated and by whom.

То	be	filled	out	by	Colossal	's	Head	of	Animal	Husbandr	y
				~)		~		~-			1

Signature _____ Date _____

 \Box Approved

 \Box Denied

□ Pending (see comments below)

Reviewers Comments:

Enrichment Assessment Form

This assessment should be completed by more than one person and the number will depend on the facility, size of staff, and scheduling. At least one primary animal care technician and one supervisor should be used to evaluate the enrichment plan. The completed assessment should be uploaded into AnimalCare® Software and emailed to Colossal's Head of Animal Husbandry and Manager of Animal Husbandry. Enrichment plans should be evaluated yearly and if any major changes happen to the environment, group dynamic, life stage, or health of the individual or group. The natural history and individual history forms should be reviewed and updated at time of assessment.

Facility: Date: Species: Individual ID: Name: Assessors:

Form Completed by: Enrichment Being Assessed:

1. What is the behavior goal of the enrichment?

2. What is the number of times/days the item has been offered? What is the length of time enrichment offered?

3. How has the animal's interaction with enrichment been observed?

4. How has the item been presented? In a group of animals? Individually? In a multispecies group?

5. If provided in a group setting, describe the species, and individuals (including age, sex, hierarchy status) that have been observed interacting with the enrichment item. If there are multiple species or taxa, provide pertinent information.

7. Is enrichment edible? If not food, how much of the item is consumed? If food, how much is consumed? Describe interaction if a food item is not consumed and describe interaction if a non-food item is consumed?

8. Has the enrichment been observed to encourage the goal behavior?

9. Has the enrichment plan encouraged behaviors besides the goal behavior? If yes, please describe the behaviors.

10. Based on observations, is the enrichment safe?

11. If the enrichment is not safe, please describe why.

12. How much time does the enrichment take to develop, implement, remove, clean, and maintain?

13. Do the benefits outweigh the costs (time and money?)

14. Would you recommend discontinuing? Not recommended? Recommend? Highly recommend? Modify the enrichment? Please explain your answer.

15. How can the effectiveness of the enrichment be improved?

Additional Comments:

]	Dire Wolf Behav	vioral Husbandry	y Shaping Plan	L					
Animal Name/ ID			Name of Behavior	r					
Species									
				- 10 ⁻¹					
Primary Trainer(s):	Primary Trainer(s):								
Foundation Behaviors	Needed:			Date Goal Reached:					
Bridge:		Reinforcer(s) and Sch Reinforcment	edules of						
Training Locations:		Additional Supplies no	eeded:						
Safety Considerations:									
Reviewed By:									
	Denied	□Revise	□Resubmit	□See notes					
Notes/Comments:									

Approximations (A)								
Throug approxin Additiona	Throughout the training process, keep in mind the approximation rule: The animal is ready to move to the next approximation in the training plan only after they have performed a behavior correctly a <u>minimum</u> of three times. Additionally, the animal needs to be comfortable and reliably demonstrate the behavior defined at each step prior to moving to the next one.							
А								
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Behavioral Expectations (Companion to Behavioral Husbandry Policy)

An addition to Behavioral Husbandry Policies and Procedures, this document is designed to give species specific direction, while the policies and procedures outline how and why these goals should be reached.

It is expected that the Colossal dire wolf will exhibit general and social behaviors similar to the grey wolf (*Canis lupus*). In the wild, wolves engage in a wide range of behaviors that contribute to their social structure and survival strategies.

Normal to optimal behaviors for developing and living within a pack structure include:

- Communicating with one another through body language and vocalizations, like howling.
- Regular use of scent marking on trees, rocks, and other objects.
- Social grooming, nuzzling, and play amongst pack members.
- Cooperative feeding.
- Creating a balanced and stable pack hierarchy.
- Strong social bonds amongst pack members.
- Successful breeding and reproduction (if determined appropriate and desired in the management plan).
- Hunting behavior. Environmental enrichment and feeding strategies will be employed to allow the wolves to display coordination, problem solving, and physical activity similar to those used in hunting cooperatively for prey items.

Wolves in the wild travel for many miles while hunting. In managed care, training and enrichment can also be used to ensure the wolves are moving around their habitats to maintain their physical health.

Signs of stress or poor health may include:

- Pacing
- Spinning or twirling
- Scaling/jumping on fences
- Increase in aggression or submission
- Overgrooming
- Excessive licking or chewing
- Diarrhea
- Hair loss
- Disease
- Decreased appetite and/or weight loss
- Reproductive failure
- Maternal neglect

If individuals or the pack are showing any of these signs, or regular conflict or heightened aggression, a lack of cohesion, or failed breeding (if determined appropriate and desired in the management plan), this would trigger further investigation, husbandry and veterinary consultation, and action to improve conditions to achieve normal and desired outcomes.

Environmental enrichment will be provided by caregivers to provide further opportunities for mental stimulation and problem-solving. This will include interactive objects like puzzle feeders, novel scents, and

objects to manipulate. Interactive objects will be approved by the animal management team and ensure that the animal is safe and engaged while using devices.

- Any enrichment will be tested to ensure the animals safety.
- Items will not be transferred between enclosures without cleaning them or ensuring they will not spread pathogens or parasites to animals in a different enclosure.
- Items will not have sharp edges or be chewed to the point of breaking off.
- Any ropes will be hung so no animal can get any part of its body caught up in it or become tangled in it.

Positive reinforcement training will also be utilized to have wolves voluntarily partake in basic medical procedures.

- When offering reinforcement to Colossal dire wolves, at no time will animal care staff use their hands or mouth to offer the food reward. Tongs that can be disinfected will be used for the safety of the animals and staff.
- Colossal recommends the use of heavy plastic meat sticks that have a round disk at the back to prevent the animal from pulling the stick into the habitat. The sticks are longer which allow staff to stand further away from the habitat.
- Tactile rewards will not be done with animal care technicians' hands unless approved by the Head of Animal Husbandry.
- Back scratchers can be used to maintain safety of animal care staff's appendages for any animal over 45 pounds.
- If the individual wolf's behavior dictates, the Head of Animal Husbandry may halt tactile reinforcement, and an alternative method may be utilized as needed to ensure the success of the training program and the safety of staff and animals.

The following list is not inclusive of all behaviors that the Colossal dire wolves can learn but is the starting point for which animal care staff can build from. All behaviors should follow the submission and approval process outlined in the <u>Behavioral Husbandry Policies and Procedures</u>.

Colossal expects the following behaviors to be trained with the dire wolf:

- Shift
- Recall to building or object
- Target
- Voluntary weighing
- Voluntary crating
- Blood draws
- Vaccines
- X-rays
- Ultrasounds

All dire wolves should be trained to allow for voluntary routine visual inspections. Each animal should present the following areas:

- Eyes
- Nose

- Open mouth to inspect teeth and palate
- Ears
- Paws (all four)
- Belly (animal should be standing on hind legs, with front paws up on a fence to allow for inspection of front paws)

Animal Welfare Management Policy

Colossal's animal welfare and animal management philosophies are rooted in the animal care principles called the Five Opportunities to Thrive. We will use these opportunities as a lens through which we will view our animal care goals, processes, and practices. These Opportunities are used to define Animal Welfare Strategies for each defined taxonomic group. Animal care teams will use them to define and inform our animal management, health, and welfare decisions.

Welfare is defined in this document as the collective physical, mental, and emotional states of an animal over time and is measured on a continuum from good to poor. Colossal uses a framework implemented by many of the top Zoological Institutions in the world that focuses on the Five Opportunities to Thrive:

1. Opportunity for a strategically presented, well-balanced diet: We will supply a suitable, speciesappropriate diet in a way that ensures full health and vigor, both behaviorally and physically. We will employ best practices to evaluate whether the animal(s) are being offered an appropriate diet in a way that encourages natural feeding behaviors, both physically and temporally.

2. Opportunity to self-maintain: We will supply a robust environment, including proper shelter and speciesappropriate substrates that encourage opportunities for self-maintenance. We will employ best practices to evaluate whether the animal(s) can maintain their own optimal health with appropriate environmental factors related to thermoregulation, grooming, mobility, and cognitive health.

3. Opportunity for optimal health: We will identify and address causes of injuries or disease, while providing supportive environments that prevent illness and increase the likelihood of healthy individuals. We will employ best practices to evaluate if the animal care regimens benefit from a clear line of communication between all responsible parties. We will work to ensure that roles and responsibilities are well-defined so that all parties involved are operating within their area of expertise as part of a collaborative effort.

4. Opportunity to express species-specific behavior: We will provide quality spaces and appropriate social groupings that encourage species-specific behaviors at animal-determined frequencies. We will employ best practices to evaluate whether the animal(s) can express meaningful behaviors at suitable intervals, with the goal of supplying appropriate diversity while meeting the social and developmental needs of the species. This may include, but is not limited to, fitting developmental conditions (social/cognitive), enriched social settings, complex environmental experiences, minimized maladaptive behaviors, and a strong and responsive relationship with the environment.

5. Opportunities for choice and control: We will provide conditions in which animals can exercise control and make choices to enhance the quality of their lives. We will employ best practices to evaluate if the animals' environment supports their ability to make choices related to space, and that training programs are based on the

principles of positive reinforcement. Enrichment and environmental conditions should include problem solving and opportunities for decision-making.

Animal Management vs. Animal Welfare

It is critical to identify the difference between an animal management issue and an animal welfare issue. Although it is possible that an animal management decision (acquiring animals, moving animals, reintroducing animals) could have a direct impact on animal welfare (developmental disruption, social disruption, physical impacts), the two are measured separately.

Animal management is the general term encompassing the processes or practices through which the desired state of being for an animal is achieved, and it is directly related to the inputs that caretaker's control. These inputs are not always measured scientifically if there is no direct evidence of the impact on outcomes for the animal.

Animal welfare is a scientifically measured group of outcomes that can be validated as having a positive or negative impact on the animal. Animal welfare is the incremental and comprehensive state of being of an individual animal or a group of individual animals. As viewed through the Five Opportunities to Thrive approach, animal welfare is a science. In practice, animal welfare is the way in which an animal interfaces with and processes its environment and is linked to animal management strategies.

Assessment of Welfare

This document describes how welfare assessment is undertaken at Colossal through a combination of daily assessments, periodic and/or annual assessments, and by event-based assessments.

Daily Monitoring

All animals in our care will be assessed daily for negative and positive welfare states as part of our routine animal care, observation, and record keeping. Each area's animal care technicians are responsible for the daily observation of all animals under their care and ensuring accurate data entry into the designated record keeping system, such as the animal care information management system, that chronicles significant events that impact the animals under their care during a specific workday. If a welfare concern is noted by the animal care technicians, then action is prompted to resolve the welfare concern by them if they are authorized to do so. If the concern cannot be easily addressed without further support, it will be brought to the attention of the Head of Animal Husbandry, and/or Manager of Animal Husbandry in a timely manner.

Animal Records System Welfare Module

The Welfare Module is a helpful tool for the monitoring of animal welfare. Teams can create their own templates to track data that they feel will be useful for the records of their animals, while not necessarily indicating status of welfare.

Periodic/Annual Welfare Assessment Process

Welfare Assessment Tool

The Welfare Assessment Tool was developed by modeling examples from zoological facilities accredited by the American Zoo and Aquarium Association and members of the World Association of Zoos and Aquariums including the San Diego Zoo Wildlife Alliance, the North Carolina Zoological Park, and the Dallas Zoo. This tool was designed to focus on measuring outputs from the animals being assessed as well as inputs provided to the animals.

The Welfare Assessment Tool is scored by placing the number associated with the answer in the column below the appropriate answer. The WAT then adds up the totals. The total in each column can be compared to the key. Please see the Documentation and Resulting Action section below for more direction in utilizing the results of the <u>Animal Welfare Assessment Tool</u>.

Welfare Assessment Teams

Welfare assessments will be completed by two-person teams that are composed of an animal care technician that directly provides husbandry care for the subject of the assessment, and a representative from the husbandry leadership team. The welfare assessment team will review relevant records, discuss the care and management history over the past 12 months with the animal care technician assigned to the team, and view the animal(s) as well as its habitat and holding and handling areas. A draft assessment form is then completed, and a meeting is held with the animal care team. Following input from everyone involved, the welfare assessment can be revised before being formally submitted.

Welfare Assessment Units

Welfare assessment units are typically groups of animals that are the same species or similar and housed together in the same or nearby enclosures, while also being under the care of the section. Our Welfare Assessment tool is designed as such that individuals with welfare concerns can be noted and flagged for indepth, individual assessment. Although an assessment may be performed on a group of animals, each animal will be considered individually when answering specific assessment questions about welfare concerns and major health events in the last year. Some mixed species communities housed together within one habitat space can be assessed as a group unit, even if multiple species are present if this has been approved by the Animal Welfare Working Group (AWWG).

Time Period of Assessments

When conducting welfare assessments, the assessment team can use their knowledge of the inputs, outputs and relevant records that exist within the past 12 months to incorporate any seasonal or social variation that could affect the welfare of the animal. In some cases, a second assessment may be deemed necessary to ascertain a more accurate representation of the animal's welfare if that animal experiences significant differences in their management and/or holding spaces between seasons. This will be noted on the Welfare Assessment Tool.

Documentation and Resulting Action

A completed welfare assessment is submitted to The Manager of Animal Husbandry who collates the results in a spreadsheet for review by the Animal Welfare Working Group at the next quarterly meeting. If the assessment team feels there is an urgent concern, or if the score exceeds an acceptable limit (> 84), the Head of Animal Husbandry should be informed, and the assessment will be submitted into the Animal Welfare Concern Process. All welfare concerns submitted within a three-month period will be reviewed by the Animal Welfare Working Group and will be ranked in order of priority for resources to be allocated.

Animal Welfare Working Group Review

The Animal Welfare Working Group will meet every three months to review all welfare assessments submitted in that time since the last meeting and recommend actions and prioritization of available resources. Once a baseline review has been achieved for all collection animals, the AWWG can make recommendations for budget priorities based on assessment results.

Event-Based Welfare Assessment

An event-based welfare assessment is initiated based on events that could significantly impact animal welfare (e.g. intense weather, change in housing, noise intrusion, construction, introductions, breeding, research, changes to normal operations, etc.). As event-based welfare assessments may be proactive or reactive, the alert for the need to conduct such an assessment can be identified by the section animal care technician and concerns should be relayed to the Head of Animal Husbandry, along with the husbandry team.

The specific welfare indicators to be evaluated are determined by:

- 1. Perceived concern for the animal(s) involved, as designated by appropriate animal husbandry leader
- 2. Available resources and staff time for more in-depth assessments involving many welfare indicators

Documentation of Results and Any Related Action

Specific welfare indicators and the frequencies at which they are monitored determine how results are documented. Documentation can either be through animal care technicians' data entry into the animal records system, or summaries of results of behavioral and/or physiological indicators monitored by the veterinary or animal curatorial/supervisor teams, which will be reported in the regular Animal Welfare Working Group meetings. Any changes in management due to the results of the event-based welfare assessment will also be discussed in the next scheduled Animal Welfare Working Group meeting.

Quality of Life Assessments

When there are concerns over the welfare or quality of life of an animal based on either medical or age-related complications, we will use the quality of life assessment tool. The AWWG will assign an assessment team to perform assessments on a regular basis, as determined by the AWWG. When the AWWG determines that welfare is at a concerning level, they can make recommendations to consider euthanasia. **If an acute event occurs in which the veterinarians and/or the Head of Animal Husbandry determine that an animal should be euthanized, then that process will occur independently of the Animal Welfare Assessments. The <u>Baseline Quality of Life Assessment Tool</u> may be used in that situation independently of the Animal Welfare Assessment to determine an animal's quality of life. Because a quality of life assessment is a snapshot of the animals overall quality of life, it should be used in conjunction with and compared to the animals' welfare assessments that are taken regularly. The Head of Animal Husbandry will utilize all these resources when determining any outcomes from the quality of life assessments.**

Animal Welfare Concern Policy

Reporting Animal Welfare Concerns

Concerns about animal welfare can be submitted by any employee and submitted through a written <u>Animal</u> <u>Welfare Concern Form</u> or email sent to Colossal Biosciences Human Resources office.

Reviewing Animal Welfare Concerns

The AWWG is committed to providing a trusted system for all animal welfare concerns, expressed from all levels of the organization. When considering an animal welfare concern, the AWWG will:

1. Involve all stakeholders in concerns affecting their areas.

2. When necessary, expect that the chain of command be utilized as the first option for reporting an animal welfare concern.

3. Respect requests for anonymity. If the requestor desires to remain anonymous, the AWWG will protect that wish.

a. Colossal has a strict policy regarding retaliation; in no way will a staff member of Colossal be retaliated against for submitting an animal welfare concern.

Animal Welfare Concern Forms can be submitted to Human Resources, Head of Animal Husbandry, or Manager of Animal Husbandry. Receipt of the Animal Welfare Concern Form should be acknowledged within 72 hours. A member of the AWWG will review the concern and a meeting will be scheduled with appropriate staff to address the concern. The AWWG will investigate the concern and determine if it needs to be investigated further. The AWWG can request that an additional Animal Welfare Assessment be completed, as well as any additional investigation necessary. Continuous updates on the status of the concerns will be communicated during the investigation process.

Animal Welfare Working Group

Composition of Animal Welfare Working Group (AWWG):

- Colossal Chief Animal Officer
- Colossal Husbandry Team
 - o Head of Animal Husbandry
 - Manager of Animal Husbandry
 - Manager of Small Animal Care
- Colossal Core Biology Team
 - o A scientist will be asked to join when appropriate concerns arise
- Animal Health
 - A veterinarian will be asked to join when appropriate concerns arise

Roles and expectations of AWWG members: The Animal Welfare Working Group members may contract outside assessors for each welfare assessment completed and then meet every 4 months to review the results. Senior staff members of the Animal Welfare Working Group (Directors, Curators, Veterinarians, Zoological Managers) will serve as permanent members of the AWWG.

Responsibilities of the AWWG

The AWWG will use its influence to develop a culture in which animal welfare is the top priority. The AWWG will use an evidence-based approach to investigate concerns about animal welfare and make recommendations to address the concerns, as appropriate.

Appendices

- A. Welfare Assessment Tool
- B. Baseline Quality of Life Assessment Tool



Animal Welfare Assessment Tool Jan :



Animal Welfare Concern Form									
Name	Date:	Time:	AM PM						
Anonymous 🗆	Address:								
Phone:									
Email	City State, Zip:								
Animal(s) Involved With Concern									
Animal(s) include name, ID:	Location: include facility	, builidng or habitat:							
Type of Animal Welfare Concern									
Social Housing Health Managemen	t 📋 Housing	U Other							
Behavioral I Nutrition I Security	Habitat	ded to fully evoluin the	000000						
Please attach any photos or documentation if available	additional pages need	ava to runy explain the	concern.						
Forms may be turned in in-person, via email, or by	/ mail to:								
Animal Welfare Working Group C/O Colossal Bioscie	ences								
3309 Elm Street, Suite 130, Dallas TX 75226									
Email: animalwelfare@colossal.com (Subject Line:)	Welfare Concern)								
A response will be provided to the email or address pr	ovided after the investigat	ion is complete							
Internal Use Only									
Head of Animal Husbandry Signature		Date							
Chief Animal Officer Signature		Date							

Tool last updat	ed on: July, 1	0 2024		Date ne	xt update is du	e: July, 10 2025
Date of Assessment:	netato5285			and a second		
Who is Assessing? Bot	th assessors s	hould be listed:				
Is this an individual or	group?					
Animal(s) being assess	ed: Include n	ames and ID nu	imbers.			
Event Dictating Asses	sment:					
Place number indicated	l in box below	v the chosen an	swer. If using t	he excel versi	on of this form	it will total answers automatically.
Only one answer per q	uestion.		od al.			1043
Nutrition	Acceptable =1	Questionable =2	Unacceptable =3	N/A=1	Not Assessed=1	Notes
Is each animal provided a species/individual appropriate diet? Does the animal have access to clean, fresh water at all times?						
Is the diet provided in a way to encourage species appropriate feeding behavior?						
Are feeding methods safe for animals and staff.						
Is there evidence that all animals are adequately consuming food.						
Are diets documented and evaluated regularly with changes implemented as needed?						
Staff members have an understanding of nutrition and safe food handling guidelines specific for the species (e.g., USDA fish/meat/whole prey thawing and handling guidelines, live feeder insect SOPs, dangerous animal feeding policy, etc.).						

Environment	Acceptable =1	Questionable =2	Unacceptable =3	N/A=1	Not Assessed=1	Notes
Habitat provides space for animal(s) to use in an appropriate species-specific manner.						
Habitat provides substrate for animal(s) to use in an appropriate species- specific manner.						
Habitat provides furniture for animal(s) to use in an appropriate species- specific manner.						
Habitat provides shelter from the elements for animal(s) to use in an appropriate species- specific manner.						
Holding provides space for animal(s) to use in an appropriate species-specific manner.						
Holding provides substrate for animal(s) to use in an appropriate species- specific manner.						
Holding provides furniture for animal(s) to use in an appropriate species- specific manner.						
Holding provides shelter from the elements for animal(s) to use in an appropriate species- specific manner.						

There is evidence of			
the animal using			
habitat space in an			
appropriate species-			
specific manner.			
There is evidence of			
the animal using			
habitat substrate in an			
appropriate species-			
specific manner.			
-			
There is evidence of			
the animal using			
habitat furniture in an			
appropriate species-			
specific manner.			
8.028			
There is evidence of			
the animal using			
habitat shelter in an			
appropriate species-			
specific manner.			
There is evidence of			
the animal using			
holding space in a			
species- appropriate			
manner.			
There is evidence of			
the animal using			
holding substrate in a			
species- appropriate			
manner.			
There is evidence of			
the animal using			
holding furniture in a			
species- appropriate			
manner.			
There is evidence of			
the animal using			
shelters in the holding			
space in a			
species-			
appropriate manner.			
Standard Operating			
Procedures for			
cleaning follows best			
practices			
The temperature			
parameters are			
appropriate for the			
species.			

The humidity				
parameters are				
appropriate for the				
species				
The ventilation				
parameters are				
appropriate for the				
species.				
The lighting				
parameters are				
appropriate for the				
species				
The noise level	-			
narameters are				
appropriate for the				
appropriate for the				
The weter quelity				
The water quality				
parameters are				
appropriate for the				
species.		 	 	
The drainage				
parameters are				
appropriate for the				
species.				
The environmental				
disturbance				
parameters are				
appropriate for the				
species.				
Environment provides				
opportunity for				
animal management.				
0-10				
Environment provides				
opportunity for				
veterinary procedures				
for the animal.				
Environment provides				
opportunity for				
emergency shifting of				
the animal.				
Environment provides				
opportunity for				
behavioral				
management of the				
animal				

Adequate reserve/isolation space is available for this animal if needed.						
Standard Operating Procedures are documented and followed by staff for the species.						
The environment is safe for the animal.						
Health	Acceptable =1	Questionable =2	Unacceptable =3	N/A=1	Not Assessed=1	Notes
Animal's weight/BCS is within normal range.						
Animal's outward physical appearance is species and seasonally appropriate.						
Animal's movement patterns are species- appropriate						
Fecal form and output and urinary appearance and output are species- appropriate.						
Evidence of rapid and appropriate diagnoses and treatment of disease and injury.						
The Quality of Life discussion is at an appropriate stage for this animal.						
Equipment/expertise to capture/restrain are species- appropriate.						
Observations of conditions and health are made and recorded in a timely manner.						
Behavior	Acceptable =1	Questionable =2	Unacceptable =3	N/A=1	Not Assessed=1	Notes
---	------------------	--------------------	--------------------	-------	-------------------	-------
Evidence of natural curiosity and exploratory behavior.						
Evidence of appropriate feeding behavior.						
Evidence of appropriate social behaviors.						
Evidence of appropriate reproductive behaviors.						
Evidence of typical sleep and resting behaviors and patterns.						
Evidence of appropriate frequency and diversity of natural behavior for this individual/group.						
Animal does not exhibit self destructive or abnormal behaviors (e.g., stereotypy).						
Animal does not exhibit abnormal or excessive aggression.						
Staff members have an understanding of species' natural history and individual history and apply that knowledge in animal care.						
Evidence of species- appropriate use of behavior goal based enrichment.						

Animal is motivated through positively reinforcing opportunities to cooperate in its daily and medical care, whenever possible.						
Choice and Control	Acceptable =1	Questionable =2	Unacceptable =3	N/A=1	Not Assessed=1	Notes
Evidence that animal chooses to participate in training sessions/program.						
Evidence that animal has ability to adapt to environmental stressors.						
Does the animal have the choice to socially interact in a species appropriate manner given its habitat?						
Evidence that the animal is provided with environmental change.						
Evidence that the animal is provided with challenges.						
Evidence that the animal is provided with coping mechanisms.						
Evidence that the animal has opportunities for choice/control in species appropriate manner:						
Evidence that the animal has opportunities for choice/control in species appropriate manner for social settings						

Evidence that the						
animal has						
opportunities for						
choice/control in						
species appropriate						
manner avoidance of						
conspecifics						
Evidence that the						
animal has						
opportunities for						
choice/control in						
species appropriate						
manner avoidance of						
interspecific						
interactions						
Evidence that the						
animal has						
opportunities for						
cnoice/control in						
species appropriate						
manner avoidance of						
guests		0	ment of the second second second			
Affective Mental State	Acceptable =1	Questionable =2	Unacceptable =3	N/A=1	Not Assessed=1	Notes
Is the animal					1100000000	
demonstrating						
behaviors that are						
strong indicators of						
positive welfare?						
No evidence of						
negative welfare						
indicators.						
	Yes	No				
Does this						
animal/group require						
immediate attention	YES	NO				
of the AWWG?						
	Add all 11	Add All Dain	Add all 21a in	Add all NI/A	Add all Not	
	in the	Auu Ali 2 S In the	the	in the	Assessed in	
Cumulativa Sooras	appropriate	appropriate	appropriate	appropriate	the	Notas
Culturative Scores.	subsection	subsection	subsection	subsection	appropriate	INOICS
	below	below	below	below	subsection	
	OCIOW	0010 W	0010 W	0010 W	below	
Nutrition	0					
Environment	0					
Environmental						
Parameters	0					
Debaujor	0					
Chains and Cantral	0					
Affective Montal	0					
Anecuve Mental	0					
Sub totals						
Sub-totals		1		1		

.....

Total		
	0	Add all Acceptable, Questionable and Unacceptable totals together. N/A'a and Not Assessed
		should not be included here.
71-80 Accept	able	
80- 89 Questio	nable	
90+ Unaccept	able	
N/A Total	0	
Not Assessed Total	0	

Baseline Quality of Life Ass	essmen	t Tool	
Species:	Staff In	tials:	
Animal ID# and Name:	1		
Name:			
Date:			
Score: Yes = 1, No = 0; unless otherwise sta	ated next	to the qu	estion.
The higher the score, the higher the quality of life is at time of the assessment. Animal Welfare Assessmen	This asse nt Tool.	ssment to	ool should be used in conjunction with the
Nutrition, Food, and Wa	ter Inta	ke	
	Yes =1	No = 0	Notes:
Is the animal eating a normal amount of food, including seasonality, if warranted?			
Is the animal eager and excited for food?			
Over the course of a week, is the animal consuming a normal quantity of food on a day-to-day basis (taking into account changes based on seasonality)?			
Is the animal's weight stable or within its normal historical range?			
Will the animal still eat their favorite food/treats?			
Is the animal drinking normally?			
Physical Health	1		
Eyesight			
Does the animal appear to be visually impaired?			
Is the animal able to navigate its exhibit (1) or is it bumping into things, etc. (0)?			
Hearing			
Does the animal still appear as though they can hear?			
Does the animal startle more easily with loud noises or with a quick approach by an animal care technician?			

Respiration					
Does the animal breathe normally, easily?					
Does the animal become out of breath after light exertion?					
Does the animal have rapid breathing when in a rested state?					
Does the animal gasp for air when in a rested state?					
Does the animal make noise when breathing?					
Appearance	•				
Are the ears being held in a normal position?					
Does the animal engage in normal grooming?					
Is the fur and/or skin in good condition?					
Is the fur thinning and/or does it have bald spots?					
Is the fur matted, ungroomed, sticking up, looking oily or ragged?					
Are there wounds or sores?					
If a wound/sore is present, is it healing (1), or are there signs of infection [bad odor/discharge] (0)?					
Has the animal lost fat or muscle, making it easy to see its skeletal frame?					
Mobility	•				
Can the animal easily get up (1) or is it a struggle and takes more effort than it should (0)?					
Is the animal moving normally (1) or abnormally-slow, stiff, limp (0)?					
Can the animal still climb the rockwork/logs/ramps in its enclosure/holding?					
Are all limbs functioning (1) or are any of them dragging or lame (0)?					
Is the animal using all of its enclosure? If not, please explain in the notes section below.					
Has the animal's activity level decreased lower than seasonally warranted?					

Does the animal appear to have balance issues such as stumbling, falling over, or a head tilt?			
Behavior			
Is the night activity level the same (1) or decreased (0)? You may need to set up a night camera to answer this question.			
If night footage was recorded, what was the number of movements/location changes?			
If the animal has a training program, do they still participate in training?			
Is the animal eager and excited to investigate changes to the enclosure or enrichment?			
Is the animal more vocal/growling?			
Does the animal appear aggressive even when not approached and is this aggression a change from its normal behavior?			
Is the animal hiding more than normal?			
Has the relationship between you and the animal changed due to its current state?			
Time Manageme	nt		
Does the animal require more time to care for it due to its current state?			
Pain Managemer	nt		
Do they look uncomfortable (droopy /worried facial expression/whimpering)?			
Does the animal's current level of pain appear to affect its appetite?			
Does the animal look unsettled, unwilling to move all or part of their body?			
If medication or treatment is needed, can it be done without restraint?			
Questions for Veterin	narian		
Score: Yes = 0, No = 1; unless otherwise sta	ated next	to the qu	estion.
	YES =0	NO = 1	Notes
Does this animal have a chronic health condition? (yes=0, no=1)			
Are there any unusual lumps or growths? (yes=0, no=1)			

Can the animal's pain or medical condition be adequately managed to m an acceptable quality of life?	aintain					
Is the pain intermittent (1), or is it ongoing (0)?						
Do they look dehydrated (loose skin, separated fur, sunken eyes)? (yes=0, no=1)						
If weight loss is an issue, is the animal able to gain weight after a diet is increased? (yes=0, no=1)						
What is the animal's Body Condition Score?						
Is the animal's BCS normal?						
	Total					
Notes Section:						

Capture, Restraint, and On-site Transportation

The nature of working with animals is unpredictable and can change rapidly. Exotic animals can present challenges when trying to physically handle them. With the diversity of species, the multiple locations and facilities Colossal Biosciences works with, and the unpredictable nature of animals, no single document can be developed to cover all possibilities, or handling situations. This document will cover the general policy for chemical and physical restraint and transportation of Colossal animals. Additional policies and practices may be applied based on the specific local, state, and federal government requirements.

Colossal dire wolves over 45 pounds are to be managed in a protected contact environment. Dire wolves should be trained through operant conditioning to enter a crate or den where they can be pole-syringed or injected, depending on their level of training at the time. If wolves are not conditioned to enter a den, they can be lured with treats and locked in. In case of an emergency, or the need to capture or handle a dire wolf, senior management at Colossal and the veterinarian will determine the best course of action in capture and handling. Never enter a space that is shared with or occupied by a dire wolf over 45 pounds (excluding domestic dogs) that is weaned, unless approved to do so. Exceptions to these guidelines can only be made by the Chief Animal Officer after an appropriate risk assessment occurs.

For this document, we will use the following definitions to describe the four types of restraint which can be used when deciding how to best provide health assessments, husbandry procedures, medical examinations, animal transports, and emergencies.

Chemical: The use of a wide variety of medications that are injected or administered for the purpose of sedation or anesthesia.

• **Example:** The use of a tranquilizing dart to anesthetize an animal.

Physical: The use of physical effort to restrain an animal.

• Example: Capturing an animal in a net or in the hands of a human handler.

Mechanical: The use of specially designed equipment or devices to safely restrain animals. If the animal is comfortable with the device, it will usually willingly enter the device and accept restraint. If the animal is not familiar with the device, it may need to be coaxed or directed into the device.

• **Examples:** Crates or chutes that an animal enters daily for a reward, or to move between areas in its habitat. Squeeze cage or other chute area that limits the animal's mobility and allows for ease of safe access to the animal.

Psychological: The use of the animal's natural behaviors and movement to position or direct it for capture and restraint.

• **Example**: Using large flat pads or blankets in addition to slowly moving toward a small ungulate species who like to hide and tuck down in dark small areas to avoid notice from predators. The pads slowly decrease the space, and the animal will go into the corner and lay down sternally. Handlers will lay a soft blanket or pad on the animal to make the animal think it can't stand up; the animal will stay calm and usually will not need full physical restraint.

Determining Type of Restraint

The species social nature and pack/herd/flock dynamics should be considered when determining what type of restraint to use. The attending veterinarian and Head of Animal Husbandry will determine the best type of

restraint to use on a case-by-case basis. Restraint of any kind where an individual animal is removed from its group, smells differently, or is missing from the group for even a small amount of time in some species can cause major behavioral and social issues that can lead to aggression and injury when the individual is returned to its social group.

In the case of Colossal dire wolves, removing an individual dire wolf can cause issues in the hierarchy of the pack, leading to increased aggression and injuries to members of the pack. Even the short-term removal can lead to fighting among the remaining pack members during the time the individual is gone, regardless of their status in the pack. If choosing chemical restraint, options to consider are immobilizing the individual in a safe and protected area in view of the pack, immobilizing the entire pack at the same time, or as close to the same time as possible, and recovering the individuals separately and returning the individuals back to their habitat at the same time. Physical restraint can also be used if deemed necessary and approved by the Head of Animal Husbandry and can be done in a safe, protected area in view of the pack. No matter which form of restraint is selected, care should be taken to limit the amount of time pack animals are removed from each other.

Whenever possible, animals will be trained with positive reinforcement to voluntarily enter mechanical restraint devices and voluntarily participate in minor medical and physical examinations, such as blood draws, eye exams, injections, and x-rays. For procedures or examinations that may cause any degree of pain or discomfort, an analgesic should be employed, and chemical restraint may be necessary. When deciding on the procedure and methods, the principle of least force should be used.

The comfort and safety of the animal and safety of the staff involved, should always be considered when determining which method of handling and/or restraint to utilize.

When using physical restraint equipment such as nets, y-poles, and ropes, the equipment must be the proper size and fit for the species and the size/weight of the individual animal and the situation. Appropriate PPE must be worn by any staff member involved with the procedure.

During animal transport, regardless of whether it's inside the facility or between facilities, proper secondary containment for the species involved must be maintained. This requires that these animals be transported inside of a covered vehicle as the secondary barrier. To prevent animal escapes, injury to the animal, staff, other species, or conspecifics, transfer in an open vehicle is not permitted unless absolutely necessary due to an extreme emergency. This is very important with injurious wildlife species such as meerkats and dangerous animals such as large carnivores. The transport vehicle and crates must be properly cleaned and disinfected after every use in ordinance with the <u>Disinfection and Cleaning SOP</u>.

Chemical Restraint

In emergency or safety situations in which immediate chemical immobilization or sedation of an animal is needed, the supervisor and veterinarian will work together to coordinate the procedure with the appropriate staff members based on the situation. The Head of Animal Husbandry should be made aware of the situation immediately. Non-emergency chemical immobilization should be planned with the veterinarian, Head of Animal Husbandry, and the animal care team. All staff involved with the procedure will be identified, notified, and prepped for the procedure in advance.

Chemical restraint will be administered under the direction of the prescribing veterinarian and/or a senior person trained in chemical immobilizations. An animal care technician trained in darting or administering the chemical agents being used will deploy the agents directly. In some cases where animals have been trained to

participate in injections, a staff member may deploy the agent as directed by the veterinarian. This may include oral medications or injections. In situations where darting is needed, the person most proficient at darting should be utilized to dart the animal. A veterinarian should be on premises and aware of the situation, be overseeing the procedure, and responsible for monitoring the anesthetic and providing the support equipment. The exception to this would be an animal escape in which the assigned manager will decide based on the situation. These guidelines provide more information on roles during an animal escape.

Chemical restraint equipment will depend on the location, species, and situation. The following may be used:

- Hand injection
- Pole syringe
- Dart gun
- Oral syringe
- Food item
- Blow pipe
- Incubation chamber
- Face mask for inhalation of chemical immobilization agents

Once anesthetized, endotracheal tubes and IV drips may be used to prolong and control the chemical restraint of the individual.

A husbandry team member will always be involved to help with proper positioning and holding of the individual animal based on the species. For example, ruminants' species must maintain a head up posture to avoid regurgitation and aspiration. This also provides a person to be responsible for maintaining the safety of the staff working around horns and hooves of ungulate species and the teeth and claws of carnivore species.

Physical Restraint

Depending on the species, size of the individual, behavior of the individual, location, training and skill of the staff available, and the situation, animals may be physically restrained or captured. No dire wolves weighing 45 pounds or more should be physically restrained without the use of another restraint method such as chemical or mechanical restraint. Like any physical restraint, it should be planned and coordinated to have the most effective staff available for the physical restraint, with emergency situations being the exception. Physical restraint poses risk to staff and animals. PPE must be worn and utilized. The PPE will depend on the species, location, and situation. At a minimum, staff should wear long sleeves, long pants, closed-toe shoes, and animal handling gloves with bite protection.

Appropriate physical and mechanical restraint equipment will depend on species, situation, and location. Staff must be trained in physical restraint, as well as the use of any device, before participating and using the device with an animal. A record of the staff training on the devices should be kept and added to the animal records management system. Staff should be trained on ways of restraint when trained with that species and at least yearly afterwards. All devices used should be made to handle the species they are being used to contain. The equipment should be tested and maintained monthly, and before it is used, to ensure the safety and welfare of the animal and staff. Crates should be adjusted or made to fit the species and the size of the individual animals, with attention paid to the life stage and medical condition of the individual animal.

Small to Medium Dire Wolf: Under 45 pounds

Squeeze cage, crates, nets, y-poles, eye covers, muzzles, or adjustable sized straps, towels, leather gloves, shields, baffles

Large Dire Wolf: Colossal Biosciences defines a large dire wolf as any dire wolf over 45 pounds regardless of age. These animals have the potential and have evolved to stalk, take down, and kill. These animals, regardless of being mother reared or hand reared, are extremely dangerous and should be treated as such. No person, including any staff members and animal care technicians, shall enter any space shared with or occupied by these animals. Only specially trained staff members may enter and work with these species. Once a large dire wolf is tranquilized with adequate amounts of a chemical immobilizing agent, staff can enter the shared space, enclosure or holding space with the animal. Prior to any procedure with a large dire wolf, all staff that are to be involved in the procedure should be identified, made aware of their role in the plan and the entirety of the plan, and any risks, equipment, and chemicals being used. A debrief should be done with all staff involved after the procedure and once the animal is safe, secure, and stable. The debrief should include successes, information learned, next steps, and areas for improvement. Both pre-procedure meeting notes, procedure notes, and debriefing notes should be recorded and placed in the individual animals record in the animal care information management system. The records should also be sent to Colossal Husbandry and Vet teams.

Exceptions to these guidelines can only be made by the Chief Animal Officer after an appropriate risk assessment occurs.

Surrogate Dog Pregnancy Diagnostic, Monitoring, and Interventional Plan

For the purpose of this document the following are defined as:

Surrogate Dog is the domestic dog that has undergone an embryo transfer and is carrying the developing embryos to term. She will give birth to the pups or undergo a C-section.

Foster Dog is a domestic dog that did not carry the embryos but may be employed to nurse the pups that she did not carry in her womb but is confirmed to be lactating.

Prior to Shipment of Domestic Dogs

The Attending Veterinarian (AV) and support staff will have confirmed pregnancy after embryo transfer by several means:

1. Repeated blood testing is performed on dogs in pro-estrous to detect a surge in Luteinizing Hormone (LH) to time a subsequent embryo transfer. The AV will then perform transabdominal ultrasound without sedation with gentle restraint at day 18, 21 and 23 days post ET to confirm pregnancy.

2. Abdominal ultrasound examinations are performed to detect embryonic vesicles as early as day 19 post-ET, and confirmed at day 21, with fetal development and heart detectable by day 28 post-ET.

3. As the date of conception from the ET is known, the date of parturition is predictable but may vary between 60-63 days post-ET. The ET and predicted parturition dates will be included as part of the complete medical records provided by the AV with the surrogate dogs.

After Arrival of Domestic Dogs

The Attending Veterinarian will perform physical exams and the following diagnostics tests with assistance from approved personnel to assess the health and pregnancy status of dogs after arrival:

- A full physical exam with bloodwork, fecal, and urinalysis will be performed within the first week of arrival to establish physiologic baselines for the dogs, during which time additional imaging studies will be performed below.
- Abdominal ultrasound examinations may be performed with gentle restraint of domestic dogs by qualified personnel to follow pregnancy progression: upon the first week of arrival, and during subsequent weeks (5, 6, 7, and 8). During the periparturient 3-day period, an abdominal ultrasound may be performed daily and repeatedly within 24 hours under veterinary discretion to track progress and fetal viability.
- Abdominal radiographs will be taken with up to 3 views, using gentle restraint at 7 and 8 weeks to visualize ossified fetal skeletons and confirm litter size by 45 days post-LH surge.
- TPR and weights will be monitored 3 times per week, upon arrival and then daily during weeks 7 and 8 through parturition, and more frequently under veterinary discretion.
- Expect signs of labor 56-59 days gestation (days post nuclear transfer). Signs start with mammary filling generally at least a few days before labor, milk able to be expressed a couple of days before labor. Dams often start digging and nesting and go off wet food within 12 hours of labor with relaxation of tissues starting around that time (evidenced by a 'dropped abdomen' appearance with thinner dorsal flanks/ topline and looking like the weight is being carried more ventrally). Signs of early labor that indicate

ready for c-section are licking, vaginal discharge/fluid, early contractions. We can monitor rectal temperature but haven't found a useful correlation.

Labor Plan/Monitoring Pregnant Dogs for Dystocia

Begin monitoring, as soon as Stage 1 labor begins (dam is restless, agitated); no external contractions are visible. Monitor fetal heart rates and progression of labor to Stage 2 in 12-24 hours. If Stage 2 labor begins in a timely manner, normal delivery should occur (typically within 1 hour). If Stage 2 labor is delayed (no delivery of a puppy and/or >2h between puppies without active contractions), or uterine contractions are weak/ infrequent, monitor fetal heart rates and evaluate condition of dam:

- Assess for potential causes of obstruction such as fetal-dam birth canal mismatch secondary to fetal oversize, dam pelvic abnormalities, vaginal canal masses/strictures, etc.
- Abdominal radiographs and/or vaginal canal exam are helpful
- Minimal database: PCV/TS, electrolytes, glucose and iCa. Treat any abnormalities detected such as hypoglycemia or hypocalcemia that can contribute to secondary dystocia.
- Perform abdominal radiographs and transabdominal ultrasound as above, to assess the position and viability of fetuses within the uterus.
- If primary dystocia is suspected or fetal distress is determined, transfer dam to on-site veterinary hospital for further assessment/monitoring and surgical intervention (See C-section below).

C-Section Recommendations

The following is the recommended plan for whelping a surrogate dog. Wait until the surrogate dog demonstrates signs of labor and perform a cesarean section.

- Perform cesarean section on-site where the dam lives; on-site will greatly reduce stress level for the dam and therefore the pups and increase likelihood of dam accepting pups.
- Time from induction to removal of pups should be reduced as much as possible-recommend clip abdomen prior to induction, have all supplies prepped, etc.
- Recommended medications for cesarean section: induce with propofol only, maintain on inhalant isoflurane, after pups removed, administer single dose of injectable opioid PRN to dam, after pups removed administer injectable Carprofen to dam, once awake administer injectable Oxytocin dose, continue Carprofen orally or by injection for 2-3 days post-op.

If the dam is stable, no obstructions are present, and the fetal HR >180 but uterine contractions are weak/infrequent:

- Begin IV fluids with 2.5-5% Dextrose; administer calcium if contractions are weak (1ml 10% calcium gluconate/4.5kg SQ; can repeat q4-6h).
- Oxytocin 0.25-1U/dam 10-15 min after calcium administration; can repeat every 30-45 minutes for up to 3 doses.
- Pups should be dried, stimulated, and warmed immediately as they're removed from the abdomen and umbilical cords torn/placentas removed. Suction of any fluid from nostrils/mouth with an ear bulb syringe and/or Dee Lee mucus trap is recommended. Do not discard some of the placentas as sometimes that helps dam with pup acceptance. Time under anesthesia and cold puppies are the biggest concerns at this stage. Warm puppies safely and immediately. Perform reviving on a warming blanket such as a 3M Bair hugger, heat pads, etc. Obtain birth weight on each puppy.

- Return pups to the dam as the dam is starting to wake up with staff monitoring closely. Show or offer placentas to the dam to lick/eat and monitor the dam very closely for appropriate maternal behavior. Do all of this immediately postoperatively as long as it is safe for the pups in the dam's pen in her whelp nest with appropriate heat sources. This will be more stressful with reduced acceptance of pups if not done in her normal environment and whelp nest. Staff should assist pups to nurse on the dam (may need to gently restrain the dam to start) once pups are warm and lively. Don't feed a cold pup. Staff may need to sit with the dam around the clock until she truly accepts pups. This can happen within 1-2 hours after waking from anesthesia if she was truly starting labor at the time of c-section and hormones are in the right place or can take up to a few days if circumstances were less ideal.
- Puppy weights should be monitored once daily and any pup that isn't nursing well and/or gaining weight well may be supplemented with Esbilac puppy milk replacer. Ideally pups will be assisted to nurse more frequently on the dam, but if supplemental caloric requirements are indicated, tube feeding by trained staff is ideal as it is much safer for the pups than bottle feeding. If bottle feeding is required, make sure to feed in a natural nursing position with an appropriate pediatric puppy bottle and/or syringe with nipple set up. Administer serum from the dam subcutaneously (~12 mLs per pup) if there is a colostrum concern or any pups aren't thriving from the start.

Indications for Electing for Cesarean Section

- 1. Failed medical treatment (IV fluids + Dextrose, calcium, and up to 3 doses of Oxytocin)
- 2. Atypical vulvar discharge (uteroverdin or malodorous) is observed without production of a fetus within 15 minutes
- 3. Fetal membranes present/protruding for >15 minutes without progress
- 4. Fetal HR is persistently <180
- 5. Obstruction is present

Surrogate Dog and Dire Wolf Pup Management Plan

Space Requirements and Housing

USDA Space Requirements. PART 3 Subpart A Dogs & Cats 141 Part 3, Subpart A, § 3.6 9 CFR AWA (5-1-2023 Edition) 2. 3. PART 3 Subpart A Dogs & Cats 4. 5 Additional requirements for dogs. 1. Space. i. Each dog housed in a primary enclosure (including weaned puppies) must be provided a minimum amount of floor space, calculated as follows: Find the mathematical square of the sum of the length of the dog in inches (measured from the tip of its nose to the base of 142 Animal Welfare Act, USDA Part 3, Subpart A, § 3.6 its tail) plus 6 inches; then divide the product by 144. The calculation is: (length of dog in inches + 6) × (length of dog in inches + 6) = required floor space in square inches. Required floor space in inches/144 = required floor space in square feet. ii. Each bitch with nursing puppies must be provided with an additional amount of floor space, based on her breed and behavioral characteristics, and in accordance with generally accepted husbandry practices as determined by the attending veterinarian. If the additional amount of floor space for each nursing puppy is less than 5 percent of the minimum requirement for the bitch, such housing must be approved by the attending veterinarian. If the case of dealers and exhibitors, such housing must be approved by the Administrator.

The surrogate dog, being a medium to large sized hound, will not exceed 40 inches in length. Based on this, the length of 40" measured nose to base of tail, was the standard used to calculate the regulated space requirement.

- The calculation is: (length of dog in inches + 6) × (length of dog in inches + 6) = required floor space in square inches.
- Required floor space in inches/144 = required floor space in square feet.
- (Length of dog 40" + 6) x (length of dog 40" + 6) = 2,116 square inches of required floor space per surrogate dog.
- 2,116/144 = 14.694 square feet of required floor space per surrogate dog.
- Additional amounts of floor space per nursing pup needs to be at least 5% of the dam's required space. For up to four pups this would require an additional 2.938 square feet at .7345 sq feet per pup. Dam with four pups would require 17.628 square feet.

The facility has six stalls where surrogate dogs will be housed with young pups. Each stall exceeds USDA minimum guidelines for domestic dogs. If the number of surrogate dogs allows, the facility allows for the dam and pups to be managed using two adjacent pens that are attached by a common shift area/gate connecting the two pens. This can either be left open as one space when just one surrogate dog is present or separated into two pens when two surrogate dogs are present and appropriate to be managed next to one another. Visual barriers will be put in place utilizing a dire wolf safe material as needed based on the behaviors of dogs and pups. There are six individual pens in the building. Three pens measure 8'5 X 8'5 ³/₈" or 101"x 101.375' which is 70.73 square feet for each individual pen. Three of the pens are each measured at 8'5" x 8'2 ³/₈" or 101"x 98.375" which is 68.62 square feet for each individual pen. In the building the pens are sectioned in groups of two with a 8'5" x 8'5 ³/₈" pen next to a 8'5" x 8'2 ³/₈" pen. The total space of combining the two pens would be 140.093 square feet. Using the combined required floor space for a surrogate dog with pups calculated above at 17.628 square feet, the 71.103 square foot pen is 4.033 times larger than required. Using the combined required floor space for a surrogate dog with pups calculated above at 17.628 square feet, the 68.999 square foot pen is 3.902 times larger than required. When utilizing the combined space of the two adjoining pens of 140.093 square feet for one surrogate dog with pups is 7.947 times larger than is required by USDA. Based on these calculations, this building can house up to six surrogate dogs with young pups. Current planning is to house up to three surrogate dogs with pups at a time, if it becomes necessary to increase this number based on reproduction plans, the appropriate housing plan will be evaluated and approved by the attending veterinarian and the Head of Animal Husbandry.

See <u>Habitat and Enclosure Requirements and Descriptions</u> for additional details of this area.

Temperature and Lighting Parameters

3.2 – USDA Environmental Condition Requirements. Indoor housing facilities. a. b. c. Heating, cooling, and temperature. Indoor housing facilities for dogs and cats must be sufficiently heated and cooled when necessary to protect the dogs and cats from temperature or humidity extremes and to provide for their health and wellbeing. When dogs or cats are present, the ambient temperature in the facility must not fall below 50 °F (10 °C) for dogs and cats not acclimated to lower temperatures, for those breeds that cannot tolerate lower temperatures without stress or discomfort (such as short-haired breeds), and for sick, aged, young, or infirm dogs and cats, except as approved by the attending veterinarian. Dry bedding, solid resting boards, or other

methods of conserving body heat must be provided when temperatures are below 50 °F (10 °C). The ambient temperature must not fall below 45 °F (7.2 °C) for more than 4 consecutive hours when dogs or cats are present, and must not rise above 85 °F (29.5 °C) for more than 4 consecutive hours when dogs or cats are present.

The preceding requirements are in addition to, not in place of, all other requirements pertaining to climatic conditions in parts 2 and 3 of this chapter. Ventilation. Indoor housing facilities for dogs and cats must be sufficiently ventilated at all times when dogs or cats are present to provide for their health and well-being, and to minimize odors, drafts, ammonia levels, and moisture condensation. Ventilation must be provided by windows, vents, fans, or air conditioning. Auxiliary ventilation, such as fans, blowers, or air conditioning must be provided when the ambient temperature is 85°F (29.5 °C) or higher. The relative humidity must be maintained at a level that ensures the health and well-being of the dogs or cats housed therein, in accordance with the directions of the attending veterinarian and generally accepted professional and husbandry practices. Lighting. Indoor housing facilities for dogs and cats. Animal areas must be provided a regular diurnal lighting cycle of either natural or artificial light. Lighting must be uniformly diffused throughout animal facilities and provide sufficient illumination to aid in maintaining good housekeeping practices, adequate cleaning, adequate inspection of animals, and for the well-being of the animals. Primary enclosures must be placed so as to protect the dogs and cats from excessive light.

The facility where surrogate dogs with pups will be housed is indoors and climate controlled with an HVAC system. This keeps temperatures and humidity levels within comfortable range at all times. Ideal temperature range will be maintained between 65-75°F with an ideal temp of 72°F. The area is well lit and will be maintained to provide a regular diurnal lighting cycle.

Domestic Dog Exercise Plan

USDA Exercise Requirements. PART 3 Subpart A Dogs & Cats 141 Part 3, Subpart A, § 3.6 9 CFR AWA (5-1-2023 Edition) 2. 3. PART 3 Subpart A Dogs & Cats 4. 5 Additional requirements for dogs. 3.8 - Exercise for dogs. Dealers, exhibitors, and research facilities must develop, document, and follow an appropriate plan to provide dogs with the opportunity for exercise. In addition, the plan must be approved by the attending veterinarian. The plan must include written standard procedures to be followed in providing the opportunity for exercise. The plan must be made available to APHIS upon request, and, in the case of research facilities, to officials of any pertinent funding Federal agency. The plan, at a minimum, must comply with each of the following: a. b. Dogs housed individually. Dogs over 12 weeks of age, except bitches with litters, housed, held, or maintained by any dealer, exhibitor, or research facility, including Federal research facilities, must be provided the opportunity for exercise regularly if they are kept individually in cages, pens, or runs that provide less than two times the required floor space for that dog, as indicated by § 3.6(c) (1) of this subpart.

Based on the USDA exercise for dog standards, an exercise plan is not required based on the space of the holding pen. Exercise is required when the space is less than two times the required floor space for that dog.

- Based on calculations made above, the required floor space for a surrogate dog with pups is 17.628 square feet.
- Two times that required floor space of 17.628 is 35.256 square feet.

• The single pen space is 69.675 square feet, which is just short of 3.95 times this requirement and the combined space of two pens for one surrogate dog with pups is 139.35 square feet, 7.91 times the required space.

Colossal is committed to exceeding regulatory and industry standards/best practices for the care of animals under our responsibility.

Domestic Dog Cleaning and Cleanliness Standards

132 Animal Welfare Act, USDA Part 3, Subpart A, § 3.1 and made of materials that allow them to be readily cleaned and sanitized, or removed or replaced when worn or soiled. Interior surfaces and any surfaces that come in contact with dogs or cats must: i. ii. 2. Be free of excessive rust that prevents the required cleaning and sanitization, or that affects the structural strength of the surface; and Be free of jagged edges or sharp points that might injure the animals. Maintenance and replacement of surfaces. All surfaces must be maintained on a regular basis. Surfaces of housing facilities – including houses, dens, and other furniture-type fixtures and objects within the facility – that cannot be readily cleaned and sanitized, must be replaced when worn or soiled. 3. d. Cleaning. Hard surfaces with which the dogs or cats come in contact must be spot-cleaned daily and sanitized in accordance with § 3.11(b) of this subpart to prevent accumulation of excreta and reduce disease hazards. Floors made of dirt, absorbent bedding, sand, gravel, grass, or other similar material must be raked or spot-cleaned with sufficient frequency to ensure all animals the freedom to avoid contact with excreta. *Contaminated material must be replaced whenever this raking and spot-cleaning is not sufficient to prevent or* eliminate odors, insects, pests, or vermin infestation. All other surfaces of housing facilities must be cleaned and sanitized when necessary to satisfy generally accepted husbandry standards and practices. Sanitization may be done using any of the methods provided in § 3.11(b) (3) for primary enclosures. Water and electric power. The housing facility must have reliable electric power adequate for heating, cooling, ventilation, and lighting, and for carrying out other husbandry requirements in accordance with the regulations in this subpart. The housing facility must provide adequate running potable water for the dogs' and cats' drinking needs, for cleaning, and for carrying out other husbandry requirements.

In our facility, these standards are to be met and exceeded. Daily and weekly schedules have been developed to ensure that animal care staff deliver on these standards.

See also Servicing Standards and Disinfection and Cleaning.

Footbaths

Footbaths will be used in all Colossal animal areas at the entrance to the area, building, or yard. Footbaths are required to help prevent the spread of pathogens, including but not limited to Parvovirus. Footbaths must be changed regularly, regardless of the disinfectant that is used. They must be changed at a minimum of once a day but may need to be changed after every use. If the footbath is 10% organic material (dirt, feces, etc.), the disinfectant will inactivate and no longer be effective. If bleach is being used as a footbath it must be at least 15% bleach solution.

Whelping Pups

A whelping box will be placed in the surrogate dog's holding area several weeks before pupping. This will allow her to become familiar and comfortable with the area before it is time for whelping. When in the

whelping window based on gestation, a constant watch will be implemented. Watch will be done primarily using the camera system to not add stress to the dog. For specifics on what to watch for and veterinary intervention markers, please refer to the <u>Surrogate Dog Pregnancy Diagnostic</u>, <u>Monitoring</u>, and <u>Interventional Plan</u>. The whelping box will have adequate heat around 85°F, as puppies are not able to make their own body heat until they are 2 1/2 weeks old. We will be using the Durawhelp X-large whelping box, as pictured below.



Managing Surrogate Dam with Pups at Dire Wolf Management Facility

The dam will continue to be managed with the dire wolf pups until such time as it is determined to be appropriate and beneficial for the dog surrogate dam to be moved away from the pups as they are age appropriate to do so. Currently, this timing is planned to be no sooner than weaning at 6-8 weeks and ideally she will remain with the pups until they are at least 12 weeks of age. During this time, brief separations of pups from the dam will be done with durations going longer the older the pups get. This will ease both dam and pups towards the eventual separation date. Depending on how the pups are acclimating, the additional step of housing the dam in an adjacent enclosure allowing visual access along a common fence line may need to be implemented. The surrogate dog dam will be removed from the pups and moved to her new adopted home, by the time the pups reach six months old or at an earlier time dictated by the veterinarian and Head of Animal Husbandry.

While at the facility, the surrogate dog will be kept in accordance with these standards as well as those additional standards outlined in the husbandry manual, particularly the sections referring to domestic dogs. Where these are in conflict for the dog, the dog standards must be met for the surrogate dog and pups when they are together.

Neonate Rearing Guidelines

Parent Rearing

It is desired that the surrogate dog dam be left to rear the pups naturally. This will be the case whether the pups are born naturally or are delivered via C-section. Veterinary and/or animal care staff will be closely monitoring to ensure that pups are nursing normally, gaining weight as expected, being groomed appropriately, that there is no aggression from the dam, and that pups are in good health. Under these conditions, they will be parent reared. If there are concerns and alternative rearing needs to be considered, including foster rearing or hand rearing, this will require the consent of the Head of Animal Husbandry in consultation with the veterinarian. If hand rearing is determined to be necessary, the goal will be to continue to socialize the pups with the dam if possible, or if not, with an appropriate foster dog. For social and nutritional reasons, attempts will be made to identify a foster dog that may also be nursing and able to provide nutrition "wet nurse" to the pups, as well as socialization. Strict parameters will be developed to ensure that pups do not become overly imprinted to their caregivers.

Survival in the wild depends in part upon their ability to hide injury. Animals that are sick or injured do not usually survive. They can be predated, consumed by their parents, left behind, or ejected from the pack; therefore, animals do their best to hide symptoms of illness and injury. Animal care technicians need to be vigilant and watch for even the slightest symptoms of illness. Make note of subtle changes in appetite, stool, posture, gait, energy levels, and stamina. Communicate concerns immediately and through the correct channels and make thorough notations in the animal records. Report any suspicious changes to your supervisor and the husbandry team promptly. Be prepared to follow up if needed. Animal care technicians are responsible for making sure the information is received by their supervisors.

Passive transfer of immunity is especially important for a newborn. They normally receive the passive transfer of immunity from the colostrum provided from suckling from their dam within the first 24-48 hours of their lives. Depending on when the pups are pulled will determine whether they receive this from suckling. If a pup is pulled before suckling, the dam may be milked for her colostrum which can be bottle fed to the pups. Additionally, 3-5 cc of the dam's blood serum should be injected subcutaneously to each pup. The serum would need to be administered a few days after receiving the colostrum or twice within the first week if colostrum is not available. Please check with the veterinarian and the Husbandry team before giving any medical treatments.

Hand Rearing

The goal is to have the surrogate dog rear all pups. No pup will be pulled for hand rearing unless it has been determined that it is not receiving proper nutrition, there is some sort of medical issue requiring it to be pulled, or the dam is behaving in a way that may be detrimental to the pup. This will be determined in consultation with vets and caregivers by the Head of Animal Husbandry. Partial hand rearing should be considered if the dam is not producing enough milk for all pups, or one or more pups need additional nutrients to ensure it has the best success at thriving and growing. The Head of Animal Husbandry will make any decisions regarding the hand rearing/partial hand rearing surrogate dam and use of foster dogs for socialization of the canid pups. Each scenario will be handled on a situational basis and the Head of Animal Husbandry will communicate with the clinical site veterinarian and appropriate experts and personnel.

If it has been determined that the best option for the pups nutritionally is for them to be hand reared, then best practices in the hand rearing of canids will be followed.

- The neonate should have its temperature taken and weighed. The pup should be weighed first thing each day before feeding to track weight gain/loss. The pup should have its temperature taken after weighing to adjust the environment temperature as soon as possible. Neonates are not able to regulate their own body temperature and can become too hot or too cold easily. A pup's rectal temperature should be between 99-101°F. If a pup needs to be warmed up, wrap the pup's body (never cover the pup's head) in warm dry towels, and/or use a heat lamp, heating pad, or a hair dryer. Be mindful not to burn the pup as they cannot regulate their temperature and will not be able to move away from the heating device. If the pup is ill, a neonate, or is compromised, it may be necessary to take the pups temperature more regularly.
- Human infant nipples are suggested in the AZA Mexican Gray Wolf Husbandry Manual.
- Never microwave the formula. Heat bottle in a human bottle warmer or in warm water.
- Only warm the amount for that feeding. Do not warm more than the amount that is supposed to be offered.
- Heat formula to body temperature. The temperature of the warmed formula/milk should be 98-99° F and feel lukewarm (not hot) when tested on the inside of your forearm or with a food grade clean thermometer. Never use mercury thermometers. If formula/milk is too hot you can burn the neonate's mouth and esophagus.

Per the AZA Mexican Gray Wolf Nutrition Guidelines, the pup should receive unmodified PetAG Esbilac Liquid Milk Supplement for Puppies.

- The amount of formula fed per 24-hour period should be 20% of the individual pup's weight. If a pup gains weight, then the amount of formula for the day should be recalculated. (See example below.)
- A bottle of 1-4 oz warm sterile water can be offered to make sure the pup is suckling properly, and the bottle and nipples are working appropriately. This is safer for the pup in case of aspiration.
- Formula should only be kept for 72 hours once the can is opened. A can lid should be used to cover formula while in the refrigerator. Once the can is opened, the can must be labeled with the date and time it was opened and by who opened it.
- If formula is transferred out of the can, the vessel it is stored in must be labeled with the date and time opened, who opened it, can/ lot number and expiration date. The open can must be stored in the refrigerator.
- The can of formula can be opened and frozen into single serving sizes and stored in the freezer for up to 6 weeks. The frozen single use bags must be labeled with date and time opened, who opened it, the lot number of the can, and expiration date of the can.

Example

850-gram pup would receive 170 ml of formula for a 24-hour period.
850 X 20% = 170 grams
1 gram= 1 ml of formula.

Calculating Formula Amounts

The amount of formula should be divided equally into several feedings spread out throughout the day. The number of feedings will change as the pup grows, with more feeding at smaller amounts in the beginning. The pup will only suckle for a few minutes at a time.

****NOTE:** Formula amount should never be lower than previous day IF the neonate loses weight. ****** If the pup loses weight, notify vets and the Manager of Animal Husbandry and Head of Animal Husbandry immediately and maintain the previous day's formula amount.

• After all calculations are completed, you may round the formula amount offered to the closest whole number.

Example

170/7 feedings = 24.28 ml a feeding- to provide the correct amount 5 feedings will have 24 ml and 2 feedings will have 25 ml.

Here is an example of daily feeding times and decreasing formula amounts for the gradual weaning of a pup.

Δαρ	Amount	Number	Time	Stimulate?					
~ <u>6</u> ~	Anount	feedings		Stimulate:					
0-24 hrs.	100% Colostrum @ 20% B.W.	7	6a, 9a, 12p, 3p, 6p, 9p, 12a	Yes					
24-48 hrs.	75% colostrum 25% Esbilac Formula Pure @20% Body Weight	6-7	6a, 9a, 12p, 3p, 6p, 9p, 12a or 6a, 9a, 12p, 3p, 6p, 10p	Yes					
48-72 hrs.	50% colostrum 50% formula @ 20% B.W.	6-7	6a, 9a, 12p, 3p, 6p, 9p, 12a or 6a, 9a, 12p, 3p, 6p, 10p	Yes					
72-96 hrs.	25% Colostrum 75% formula @ 20% Body Weight	6-7	6a, 9a, 12p, 3p, 6p, 9p, 12a or 6a, 9a, 12p, 3p, 6p, 10p						
5-21 days	100% Formula @ 20% Body Weight	6	6a, 9a, 12p, 3p, 6p, 10p	yes					
3wks - 3 mo.	20 % of Body Weight Start offering formula-soaked puppy chow, increase soaked puppy chow/meat with consumption	5	6a, 9a, 12p, 3p, 6p	no					
3 - 3.5 mo.		4	7a, 11a, 2p, 6p						
3.5 - 4 mo.		3	8a, 12p, 5p						
4 - 4.5 mo.		2	8a, 5p						
4.5 - 5 mo.		1	8a						
	Weaned								

Establishing Nursing

Animal care technicians will need to assist the pup in taking the bottle. Care will need to be taken to prevent the pup from aspirating milk and the nipple should be checked daily to make sure it is not leaking or draining into the pup's mouth. The pup needs to suck the milk out of the bottle, not just have the formula/milk poured into its mouth.

When the neonate is pulled from its dam, the pup's rectal temperature should be taken. The next priority is to establish nursing. If the pup is cold below 99-101°F, it will need to be warmed up before trying to feed it. Once the pup is at a good rectal temperature of 99-101°F, nursing can start to be established. The pup's rectal temperature should be taken once a day if it is doing well. Remember to make sure that the pup is at a normal body temperature before attempting to feed.

- Warming methods can include setting the pup up in an area with heat lamps, placing the pup in a temperature controlled neonatal incubator, and/or wrapping/rubbing the pup with a blanket. Whichever method is employed, facilities must use a thermometer that can be monitored from outside the whelping box and or den area. The thermometer should be in an area that will accurately measure the temperature of areas where the pups will lay and is not in an area where it can cause harm to the dam or pups. Heat lamps must be placed far enough away that the whelping area stays within the temperature guidelines.
- Patience must be given to animals that have successfully nursed from the dam, as they are unlikely to readily take a bottle. The nipple and taste will be different, so the neonate will need to learn to take the bottle and formula. Good communication and observations between animal care technician, management, and veterinarians will help the neonate to be successful in nursing from a bottle. A finger can be placed in the mouth to determine if the animal has a suckling response.
- If there is no suckling response, and the neonate is calm, it may be beneficial to leave the area and come back in 10-20 minutes and try again.
- If the neonate is giving a suckling response, then a bottle may be offered. The bottle may be warmed in a container of warm tap water or the bottle warmer. Never microwave the formula directly.

Initially, try to feed by slipping the nipple in. Make sure the neonate's tongue is forward and in the proper nursing position. Cup your hand gently but firmly around its mouth. Jiggle the nipple back and forth slowly to help the neonate create suction.

- Even if the neonate does not nurse, it should give you an indication of its nursing response. Never squeeze formula into the pup's mouth, because the formula may be aspirated into the lungs.
- Encourage nursing behavior in a pup with physical stimulation. Touch and gently rub the neonate on its back, face, or head. This can mimic the maternal care and licking the dam would be providing the neonate. Learn the preferences of each neonate and communicate them to your teammates as consistency is the key and will help in future feedings.
- Do NOT continue to let the neonate suck air when the suckling noise changes or when the formula level gets too low.

Stimulating the Pup

Stimulating the pup should be done until the pup starts urinating and defecating on their own. To stimulate the pup, gently run a damp cotton ball, soft moistened paper towel, or soft rag over perineal and/or penile areas while the pup nurses or right after nursing if the pup requires assistance. Make sure not to overstimulate the pup as this can lead to irritation, chafing, and abrasions.

- Stimulation should occur several times a day, but not necessarily at every feeding because overstimulation can result in rectal irritation or loose stools. Discontinue stimulation when the pup is observed eliminating on its own consistently.
- Check for recent signs of urination and defecation before stimulating/feeding. If the neonate has not urinated or defecated for the last 2-3 feedings, stimulate the neonate.

- Make sure to check neonates' tail and rump area for signs of dried feces and clean off, as necessary.
- Remove any signs of defecating and urinating when they are found so accurate records can be kept. If you leave it, the next person will not be able to tell if it is new or old.
- Constipation and diarrhea are often seen in hand reared animals and on occasion they will require changes in diet. Any instances should be reported to the husbandry team immediately, so a course of action can be planned. Fecal samples should be collected in case the veterinarians need it. If a sample is not needed it can be discarded later.

Understanding Pup Behaviors

Become aware of and used to reading the pup's reactions to your approach. Typical positive signs are approaching the animal care technician, tail wagging, forward ear positioning, vocalizations, and mouth movements signaling an interest in nursing.

- Unresponsive or frightened neonates will need to be managed differently. Quality time spent getting such a neonate used to your presence should create an atmosphere of acceptance. A positive experience may be established by short sessions (5-10 minutes hourly), with only part of that time spent trying to feed.
- Know when to quit. End a session if either you or the pup becomes frustrated and let your supervisor know. Hand rearing can be rewarding and frustrating. Letting someone who is not frustrated try can sometimes mean success for the neonate, which is the most important thing to remember. The goal is to get the neonate to nurse, it does not matter who does it as long as the pup eats.
 - On occasion, you will find that the pup is close to nursing: looking for the nipple but not allowing you to slip it in. It may be advantageous to leave for 10-15 minutes and return rather than allow the animal to become too frustrated to continue.
 - Sometimes you can do all the above and not get a positive response. Double check the medical status of the neonate A dehydrated pup will become lethargic and have problems thermoregulating and a low temperature is sometimes indicative of a low blood glucose level. If this is suspected, notify the manager and the Head of Animal Husbandry immediately.

Things to Consider with Neonate Rearing

Although nursing is an instinctive behavior, some learning is involved. Even dam-reared neonates may need to learn to nurse efficiently. Therefore, as long as the neonate appears to be alert and hydrated, be persistent but patient.

Every feeding will be documented, weight should be taken daily at the beginning of the day, and amounts offered and consumed should be recorded every time. Urination, defecation and the appearance of the urine and feces should be noted on the hand rearing logs and in the daily report.

A neonatal exam will be done at a time determined by how the pups are being reared, the comfort of the dam, and how the pups appear to be doing. The timing of the neonate exam will be determined by the husbandry team and Veterinarians. The neonate exam will include a general physical exam, blood draw, cleaning of umbilicus, temperature, any supplements or vaccines, and anything else determined by the Head of Animal Husbandry.

The hand reared pup's environment should be kept around 85°F until the pup is a month old, and depending on its health, its environment may be kept at this temperature for longer. If a pup is having trouble maintaining

body temperature an incubator may be utilized to help stabilize the pup's temperature. We suggest keeping hand reared pups in an incubator until they are 10 days old, but this will all depend on many factors including but not limited to the individual pup, the environment in which the pup will be housed, if it is being raised with other pups. The Head of Animal Husbandry should be involved in all decisions regarding the pups rearing.

Weaning Off of Formula

At three weeks of age, begin to offer formula-soaked puppy chow and slowly wean the pups from the bottle. This can be done by reducing the number of feedings if the pup is gaining weight consistently and learning to eat the puppy chow. If a pup is not consuming formula, making milestones, or consistently gaining weight, the husbandry team and veterinarians must be notified immediately.

Based on the pup's development and its solid food consumption, the amount of soaked puppy chow will be replaced with dry food. This should not reduce the total amount of food; just change the way the food is presented. The pup's teeth need to be strong enough to handle dry food. The pup needs to be eating and chewing the small meat chunks and dog food. The timing of when to start the transition from soaked to less soaked to dry, will depend on the individual animal's development and a conversation with the Head of Animal Husbandry, the veterinarian, and management.

Weaning will be adjusted based on many factors including but not limited to the pup's behavior, development, age, and medical condition. These complex decisions will be made by the Head of Animal Husbandry with input from the site veterinarian and supervisory staff.

Singleton Pup

Socialization of singleton pups will be handled on a case-by-case basis by the Head of Animal Husbandry. The Head of Animal Husbandry will get input from parties involved in the rearing of the singleton dire wolf and make the best decision for that pup's welfare utilizing the resources available at the time of that canid's rearing. As many options are available to provide good socialization of singleton canid pups, this document could not attempt to cover all scenarios that could be possible outcomes and pathways to socializing and providing optimal welfare to that individual animal.



Social Dynamics, Introductions, Breeding Plans, and Contraception

Colossal dire wolves are social by nature and consideration should be made to ensure they are managed and cared for in a way that ensures animals are in a group structure that supports and meets the social, physical, and psychological wellbeing of this species, while facilitating species appropriate behaviors for Colossal dire wolf species. In the wild, dire wolves live in multigenerational packs. Pack size and dynamics vary related to the environment, resources, territory size, prey size, and availability. Social relationships within dire wolf groups are determined by age, sex, season, and hierarchy.

Dominance and Aggression Interactions

Dominance interactions between dire wolves can turn violent and even deadly. Care should be taken to balance the need for dominance behaviors as a normal and needed behavior for the dire wolves to partake in, while not allowing the interactions to escalate to extreme injury or death. Separating dire wolves that are involved in dominance behaviors can prove to escalate and prolong the interactions as the dire wolves need to work out for themselves their hierarchy in the pack. Animal care technicians should be mindful not to let the emotions of watching the fight cloud the judgment of letting the dire wolves be dire wolves and work out their social structure. Animal care technicians and supervisors should separate dire wolves if one or both animals will/could lead to critical injury or mortality. All these interactions should be recorded with the cameras in place in their enclosures. These video recordings should be put in the animal's record in The animal care information management system.

Contraception

Contraception of dire wolves has been found to be effective for managing opposite sex pairs and groups. However, long term use of contraception can have long term and irreversible effects on the dire wolves. Facilities should contact Colossal Head of Animal Husbandry to discuss options if they are having trouble managing opposite sexed dire wolves. The <u>AZA Reproduction Center</u> has tools and resources available to understand the different reversible contraception available for dire wolves. The contraception plan for Colossal dire wolves will be further developed based on the age and social dynamics of the pack and will be decided by the veterinarian and the Head of Animal Husbandry.

Same Sex Groups

Same sex groups work best if the dire wolves are siblings or have been raised together from an early age, so they grow up as a sibling pack. All male groups tend to work better than all female groups. The AZA Large Canid Husbandry Manual records that introductions of same sex groups that were not raised together has worked successfully a couple times, but is not common practice in Mexican grey wolves, and has not been successful in red wolves. It does not provide insight into timber wolves/grey wolves/coyotes specifically.

Introductions/Forming Groups

Introduction and formation of the pack should be done with close monitoring and supervision. The animals should be in a large open space, without areas where one can be cornered and/or trapped. It is helpful to have all areas open to the dire wolves to allow for areas of refuge and provide areas without open sight lines.

Introductions should be planned with on-site animal care staff and the Head of Animal Husbandry. This ensures the appropriate staff, precautions, plans, and equipment can be put in place to try and limit injuries to the animals, allow staff to develop emergency response plans, and be available to separate animals. The introduction process will be fluid and adapt to all the dire wolves' behaviors that are involved in the introduction. The introduction plan and its progression or recession should be based on the animal's behavior. Timelines should be guidelines and flexible based on the animals behaviors and not a calendar or staff schedules. A neutral place is beneficial but not required, but existing territories may slow or change the process. Introductions of same sex groups (male or female) should always be avoided during breeding season. Hormone levels for both sexes are elevated during this time and chances of severe aggression are escalated.

- There should be a howdy plan at the beginning of the process to allow the dire wolves to see/smell and get used to each other before they are able to touch each other or place appendages near fences where other dire wolves can grab hold of them. A three feet buffer zone is a good start but may need to be larger depending on the individual history, size, and demeanor of the dire wolves being introduced.
- Part of the introduction plan should allow for the dire wolves to share a fence line. This will be determined after all parties are comfortable with the dire wolves' behaviors being separated without the ability to touch each other. This process will need to have separation plans, emergency response procedures, and a veterinarian in case of any injuries. This process usually takes 1-2 weeks of monitored interactions with people nearby and ready to intervene. The dire wolves should not be left unattended during this process.
- Next, there will be a time when the individual dire wolves will take turns having access to a common space. They can still have shared fence lines, but it may be that in the morning Dire Wolf A goes into the large habitat while Dire Wolf B is in a holding pen. In the afternoon, Dire Wolf B gets access to the large habitat while Dire Wolf A is in a holding pen.
- The next step is to give the dire wolves physical access to the same space at the same time. The space should have quick and simple ways for staff to safely separate the dire wolves without staff sharing space with the dire wolves. At no time should staff share space with any Colossal dire wolf over 45 pounds, unless it is approved by the Chief Animal Officer. Staff should expect to see aggressive behaviors such as lunging, snapping, growling, chasing, biting, and wrestling. Dire wolves should be monitored during these interactions. They should be introduced during the day and separated at night for at least 1-2 weeks, but animal managers should be flexible and move forward if they are seeing positive behaviors, as consistently separating them can increase stress levels and cause more aggression and be detrimental to the introduction process. This process may be a step forward step back process.
- Once dire wolves are not showing any moderate to significant aggression they can be left together overnight. This decision will be made by the Head of Animal Husbandry. If introducing two pairs or groups, the groups should be left together and managed as two entities unless the established groups start fracturing during the introduction process, at which time the team will regroup and adjust the plan.

Considerations for the space during the introduction process: When forming groups of same sex adult animals, facilities will need to provide multiple food and water stations—more than the number of animals is a must. These food and water stations should be spread out as much as possible to prevent an animal from monopolizing and guarding the resources. There must be multiple places for refuge and hiding, along with dens

that have multiple entrances to prevent animals from being cornered. Feed and water areas should be in open spaces so animals can eat and drink without fear of being cornered.

Separating and Reintroducing Dire Wolves

When making the decision to remove, stop introductions, or separate dire wolves, caretakers should be prepared to remove dire wolves if necessary. This decision should be made with the Head of Animal Husbandry unless it is an emergency.

If an individual is temporarily removed, the remaining pack members may re-align their social structure in its absence. If/or when attempts are made to return the individual to its pack, it may be difficult to impossible to return it to his pack. The dire wolf being reintroduced to the pack may have difficulty re-establishing itself in the pack. It may lose its rank as the hierarchy has changed in its absence which can create social conflict or injury to the reintroduced member or other members. In some situations, it may be prudent to separate all members of the pack and then reintroduce the pack members at the same time. When doing annual wellness exams, it is best practice to do the whole pack on the same day to ensure that no one animal is required to be reintroduced as all animals will be anesthetized and recovered on the same day and can be placed back into the area simultaneously. This is imperative to help maintain the current social dynamics within the pack. This strategy should be considered any time a single member of a pack may need to be examined.

Lone Dire Wolf

Single animals do occur in the wild, however it is not the norm, and dire wolves are inclined to be with conspecifics. In managed care, lone dire wolves will do well but require a robust and full behavioral husbandry plan to replace their high need for social interactions. The Head of Animal Husbandry will be responsible for determining if any Colossal dire wolves require being housed and managed singly as this is not Colossal recommended way of managing Colossal dire wolves. Socialization options will be explored and exhausted to attempt to avoid managing any dire wolf as a singleton.

Neonate Rearing

Please see Neonate Rearing Guidelines

Preventative Medicine Program

Colossal prioritizes the health and welfare of its animal population and staff by using a multifaceted approach through standard operating procedures (SOPs), training, and other programs as needed. This document serves to outline the Preventative Medicine Program (PMP) for the animals in Colossal responsibility. The PMP includes quarantine, parasite surveillance and control, immunization, infection and contagious disease monitoring, nutritional evaluation, body condition scoring (BCS), husbandry oversight, and vermin control.

The Head of Animal Husbandry and the attending veterinarian are responsible for the oversight of the health and wellbeing of the individual animals and the collective health of the animal population. To ensure animal health and wellbeing, Colossal will regularly assess individual animals, animal groups, and colonies and improve upon the methods utilized to do so. In addition to the on-site veterinarian and animal care staff, a local consulting veterinarian clinic should be identified and may be used if necessary.

Methods used to assess and measure health and wellbeing include:

- Daily observations by animal care staff, animal care managers
- Daily staff morning meetings
- Weekly staff meetings with Colossal representatives
- Regularly scheduled veterinarian general assessment, will be done at least monthly
- Regularly scheduled on-site inspections by the Colossal husbandry team, at least bi-annually.
- Bi-annual welfare assessments
- Animal welfare anonymous reporting
- Shoe in/shoe out
- Footbaths at entrance to animal areas

On-site and Visual Inspections

All visual inspectionsvwill include:

- Every Colossal animal will be visually inspected by the team. Binoculars will be used to verify tags and look more closely at any animal that does not approach the inspectors.
- BCS will be given.
- Diets will be revised based on BCS that are not 4-6 on a 9-point scale.
- Medications and supplements will be reviewed.
- All aspects of each animal's physical appearance will be scrutinized which will include eyes, ears, horns, antlers, fur, feathers, scales, hooves, paws, feet, external reproductive organs, etc. Any issues noted will be followed up with the appropriate remedy which may include veterinary exam, hoof and nail trim.
- Exhibit maintenance will be reviewed and needs/concerns addressed. These needs may have already been repaired if they posed any risk to the animals. This will also provide a platform for improvement of facilities and awareness of challenges staff may have with facilities. This allows for future improvements to facilities for animal welfare, as well as animal and staff safety.
- Minor veterinary treatments may be scheduled to be performed at the monthly inspections.
- Major veterinary procedures will be planned at weekly meetings with the husbandry team and appropriate veterinarians.
- All on-site equipment will be reviewed, supplies reordered, and new equipment needs documented.
 - o New equipment needs will be discussed at time of the next weekly meeting with Colossal
- All medication will be inventoried and inspected for expiration date.
- All food items, including all diet and food enrichment, will be inspected for freshness and dating.
- Enrichment items will be inspected for wear and safety.
- All areas will be inspected for cleanliness, including storage areas.

The IACUC inspectors have their own format to comply with.

Daily Wellness Assessments

Animal care technicians are responsible for monitoring and reporting on the health and wellness of the individual animals and groups of animals in their care daily. They are required to document any and all changes

to the animal in The animal care information management system. Please see <u>Daily Health and Wellness</u> <u>Evaluations</u> for more details, fecal chart, and body condition chart.

Quarantine

New Colossal animals will be quarantined which includes a determination that animals are medically cleared before sharing space with other animals. Veterinarians and the Head of Animal Husbandry will recommend procedures based on each animal species, individual circumstance, medical history, site location, and facility infrastructure. If animals are going into empty enclosures/barns/habitats or holding areas, these areas will be considered their quarantine spaces. If the spaces have mesh walls near existing animals, the area will be modified with solid walls using plywood or plastic. Separate tools will be utilized, staff will utilize footbaths and the recommended disinfectant appropriate for the species (examples: bleach, Virkon, Rescue). Animal debris from quarantine will go into a designated location away from where other animals' debris/waste is dumped to prevent cross contamination. If possible, debris should be bagged and transported to the location. Staff may use coveralls, foot covers, change of shoes, change of clothes, or any other means recommended by the attending veterinarian and husbandry team.

Animals in quarantine will have the same daily husbandry and welfare requirements as other Colossal animals. Animals in quarantine will have two fecal runs to monitor their parasite loads, one within the first week of arrival, and the second 14-21 days later.

Dire Wolf Vaccinations and Treatments

Dire wolves are susceptible to most diseases that domestic canines are. Vaccines should be determined and scheduled with a veterinarian. Based on the AZA Mexican Gray Wolf Husbandry Manual, Colossal suggests that dire wolves receive Canine distemper, Parvovirus, Rabies, Leptospirosis, Coronavirus, and Lyme disease vaccines (if present in the area). They should also receive regular heartworm preventatives. Pups usually begin receiving their vaccines around 6-9 weeks of age and it will depend on the vaccine as to how much/what/ dosing/series are and when boosters are needed. Please consult the Head of Animal Husbandry and the attending veterinarian for timing and scheduling. See <u>Dire Wolf Preventative Medical Husbandry</u> section for more details.

Parasite Control

Fecal tests will be run on a schedule determined by the veterinarian and a senior member of the Colossal husbandry team. Fecals will be collected quarterly but may be changed based on history of the species/ individuals. The current fecal schedule is March, June, September, and December. Fecal tests will be run on any individual animal that presents with abnormal feces, starts to have poor coat quality, edema under bottom jaw, lethargy, or change in diet consumption that is not due to some other obvious reasoning. Veterinary staff and the husbandry team should be advised of the change in the individual at the time of the change and the results of the fecal testing. Fecal collecting calendars and results will be documented in the animal records management system in the animal's daily record.

Table 11: Gastrointestinal Grading Sheet for Red Wolves (adapted from Waddell 1998)

Grade 1	Greater than 2/3 of feces in a defecation are liquid. Feces have lost all form, appearing as a puddle or squirt.
Grade 2	Soft/liquid feces are an intermediate between soft and liquid feces, with equal amounts of soft and liquid feces within the defecation.
Grade 3	Greater than 2/3 of feces in a defecation are soft. The feces keep enough form to pile but have lost their firm cylindrical appearance.
Grade 4	Firm/soft feces are an intermediate between the grades of firm and soft, with equal amounts of firm and soft feces within the defecation.
Grade 5	Greater than 2/3 of feces in a defecation are firm. They have a cylindrical shape with little flattening.

When treating parasites, veterinarians and the husbandry team will formulate a deworming plan based on the species of parasite, the species that is affected by the parasite, parasite resistance in the pasture, species, and area. A follow up fecal test should be run 14-21 days after the treatment is finished. Some treatments may be a series of treatments, and the following feces should be taken 14-21 days after the last dose in the series. Fecal tests will be run no less than two times a year.

How to Collect Fecal Samples

Collecting feces for testing must come from fresh feces that the animal caretaker witnessed being produced, and it should be collected as soon as safely possible. It can be collected in a plastic bag, fecal collection cups, or other clean approved apparatus. The feces should be labeled with the species of animal it came from, the individual animal it came from, date, time, who collected, location (for example, which enclosure that animal is in), and then placed in the sample collection fridge until it is shipped for testing or run by the clinic. Group fecal samples can be collected when approved by the vet and husbandry team.

Microchips/Unique ID

Colossal dire wolves will be given a microchip placed between the shoulder blades either when they are brought into the facility or at one of the neonate/early exams. The timing will be determined by Head of Animal Husbandry and the veterinarian.

Dental Examinations

Teeth will be examined, and if deemed necessary, cleaned on all animals during any immobilizations. Routine dental examinations will be done if an individual animal is known for having dental problems. In the case of specialty species or animals with major dental problems, a dental veterinarian will be employed. This will be done at the discretion of the veterinary staff and Head of Animal Husbandry.

Zoonotic Disease

Employees working with animals will be trained on zoonotic diseases. All employees will take a yearly class/ training on zoonotic diseases. Each area will be supplied with Personal Protection Equipment (PPE) for staff to wear and utilize when working with animals. The PPE will be required and tailored to the species of animal and the risks associated with the species. PPE can include, but is not limited to vinyl exam gloves, safety goggles, face shields, surgical masks, respirators, and coveralls. The veterinarians will have a good idea of the risk and zoonotic diseases potential and will work with staff and human medical doctors in the area if a staff member is exposed to a zoonotic disease. The veterinarian inform the Head of Animal Husbandry immediately if any animal is diagnosed with a zoonotic disease. Local, state, and federal authorities will be advised of any reportable zoonotic diseases occurring at a facility.

Nutrition

Diets are reviewed several times a year at the monthly veterinarian visual inspections and at the twice yearly site visits. This will include looking at health challenges that could be associated with nutrition. Diets will be formulated and evaluated for life phases and stages, and illness. Diet changes and questions can be addressed during morning meetings, weekly meetings, and monthly visits. At any time, anyone with a concern can bring it up to their supervisors and the Colossal husbandry team. The nutritionist can and will be consulted as needed to ensure Colossal's animals receive the highest quality nutrition based on scientific and industry standards.

Necropsies

The veterinary staff must conduct necropsies and collect samples on any death of an animal. If the team is unable to perform necropsies or collect samples, they should notify the Head of Animal Husbandry in advance. Photos and results will be put into the animal's records. The veterinary staff and the Head of Animal Husbandry will work together to decide on the tests that should be run and what samples need to be collected. Samples will be sent to appropriate laboratories for further analysis if needed and results will be sent to the veterinarian and Head of Animal Husbandry, who will then send them to IACUC and other appropriate parties.

Drinking Water

Water quality assessments will be conducted annually or more frequently based on results. Action will be taken if results indicate that the water quality is not suitable for animals to drink. All areas have an automatic refilling water bowl, such as a Nelson water bowl. Colossal dire wolves should always have access to a clean water source.

Cleaning and Disinfecting

All animal areas are cleaned and sanitized on a routine schedule that is documented in The animal cart information management system. The Colossal husbandry team is responsible for maintaining and updating the schedule to ensure that all areas are maintained up to the industry professional standards. The veterinary staff will advise on the best products and dilutions to use to ensure animal health and safety.

Dawn dishwashing detergent or a similar brand is used as a cleaning agent. Sanitizing is done with 10%-15% bleach solution for food apparatus, enrichment items, and water sources. Items must be soaked in the solution for 10 minutes. Smaller food and water bowls can also be run through a commercial grade kitchen dish sanitizer. The temperature of the sanitizer must reach 180° F, and the wash cycle must be at least 110° F.

Floors and walls of stalls, dens, barns, and enclosures will be sanitized at least once a week with the use of a steam genie at a temperature of 145° F or higher. Sanitization will be done more often if animals are sick/ compromised. Veterinarians and husbandry teams will modify as needed based on the needs of the animal and

the situation. Any time animals are moved into a new location, the area must be sanitized before the new animals are introduced to the area, unless there is a need for olfactory cues to be in place for that species as is the case for certain species to breed, define territory, and maintain hierarchy. The goal of sanitizing is to maintain best practices and optimal animal health, not to negatively impact animal welfare. Some animals communicate and need olfactory cues to maintain their psychological safety.

Animal troughs and pools will be sanitized at least once a week and more often if needed based on such things as animal use, seasonality, algae growth, and debris. The schedule will be maintained and recorded the animal records management system. The trough and pools will be sanitized with bleach and rinsed with copious amounts of water.

For more information, please see Disinfection and Cleaning.

Footbaths

Footbaths will be used in all Colossal animal areas at the entrance to the area, building, or yard. Footbaths are required to help prevent the spread of pathogens, including but not limited to, Parvovirus. Footbaths must be changed regularly, regardless of the disinfectant that is used. They must be changed at a minimum of once a day but may need to be changed after every use. If the footbath is 10% organic material (dirt, feces, etc.), the disinfectant will inactivate and no longer be effective. If bleach is being used as a footbath, it must be at least 15% bleach solution.

Shoe In/Shoe Out

Animal care staff will change into a different set of shoes designated for the area to care for the Colossal dire wolves. If staff do not have a specific set of shoes designated for the area, they will utilize surgical boot covers. The designated shoes and/or boot covers will be worn in all Colossal dire wolves' areas. When staff leave the area, they will change back into their other shoes. Footbaths will be utilized as well to help eliminate any pathogens contamination to the dire wolves.

Pest Control

Rodents, insects, and other species deemed pest species, will have a pest control program that is developed with the advice of the veterinary staff. The program will be evaluated by veterinary staff and the Head of Animal Husbandry for any threat they can pose to the animals in managed care. Pesticides will be evaluated, monitored, and presented in manners that do not pose a threat to Colossal animals or non-pest species. Food storage areas should be monitored daily for pest species activity and action taken if evidence is present. Outside pest control companies may be utilized to help control pest species.

• **Bait boxes** will be used in non-animal areas to prevent pest species from entering animal barns and enclosures. Bait will be checked at least once a month, and scheduled, tracked, and managed in the animal care information management system, along with having a printed map in the building to identify where the bait boxes are. The area around the bait boxes will be monitored by staff to ensure that no bait is found outside of the box and that the boxes can never be accessed by collection animals. A map of where bait boxes are located should be posted at the entry to the room, barn, or enclosure so people can easily know where they are. This map/list should be uploaded into the animal care information management system and updated if the bait boxes are moved.

Brand Name	Chemical Name	EPA Registration #
Contrac Blox	Bromadiolone	12455-79
Tomcat with Bromethalin	Bromethalin	12455-132-3240

Bait Box Locations

Facilities should list where all bait boxes are and have a map in each area to direct people to the bait box.

- **Traps** including live traps, snap traps, and sticky traps may be utilized to keep rodents and other vermin out of collection animal areas. Traps will be checked daily. Trap locations, what was used to bait the trap, when set, and if anything was trapped will all be recorded at time of use in the animal care information management system. Colossal should be notified of the need for and use of traps in daily animal resorts and in the weekly zoom meetings.
- Fly Traps including sticky traps and fly bags, may be utilized in barns, buildings, and areas surrounding entrances to buildings to help reduce the populations. These traps can be utilized near animal waste receptacles. Fly traps should not be placed anywhere a Colossal animal can access. They should also be in places where humans do not get stuck in them.
- The species appropriate **Fly Spray** may be utilized for ungulate species and carnivore species. Before utilizing fly sprays, the veterinarian and Colossal husbandry team should be advised of the need for and type of spray being used. Fly spray should be applied based on manufacturers recommendations and dilutions. The spray should never be sprayed in the animals' eyes, ears, nose, or mouth.
- **Mosquitoes** may be controlled by fogging with foggers approved by the veterinarian and the Head of Animal Husbandry. If more control is needed based on location, approved mosquito-controlled granules can be added to areas in which the granules will not be touched or consumed by collection animals. Mosquito fish can be added to natural ponds if they do not pose an invasive species issue for local areas.

Expired Drugs

Expired drugs will be removed and placed in a marked box. The veterinarian will properly dispose of all expired drugs. The expired drugs should be found at monthly site inspections of animals. The animal management and veterinary staffare responsible for making sure inventory is current and stocked.

Waste Disposal

Animal waste and food debris should be picked up, put in a trash bag, and dumped in the appropriate dumpster for the species and conditions, and removed by a waste disposal company. Proper PPE must be worn when handling animal waste.

Hazardous Waste

Hazardous waste comes in many forms, and whether you're at work or at home, it's important to dispose of it properly to avoid doing damage to the environment or hurting someone. Use these 10 tips to avoid making unsafe choices when you dispose of hazardous waste.

- Keep It in the Right Container: Whether it's a corrosive substance like lye or a pollutant liquid like used motor oil, it's important to keep hazardous wastes in their original containers before disposal. Putting hazardous wastes inside food containers and other packaging may cause confusion and result in unsafe exposure or spills.
- Understand What Counts as Hazardous: It's important to know what is and isn't hazardous as an early step toward proper disposal. Formal definitions can be hard to follow, and if you're dealing with materials you're unsure of in a non-professional context, you might need to use your best judgment. For example, anything that's harmful or deadly if swallowed, like drain cleaner or motor oil, is considered hazardous.
- Know What Your Garbage Collector Will Take: Some garbage collection companies have websites that detail what they will and will not take. Some are willing to take a range of different hazardous materials, including medical sharps and lawn chemicals, though you might need to dispose of these things in a specific way to ensure handler safety.
- Look for Special Collection Events: Some forms of hazardous waste, such as unused medications, can pose hazards to the environment if they're disposed of in landfills. Your local community may offer special medicine collection events that allow for the safe disposal of these materials.
- **Reduce Consumption of Hazardous Substances:** If you don't deal with hazardous materials in the first place, you won't need to worry about dealing with waste. Try to find alternatives to harsh cleaners. For example, use vinegar instead of glass cleaner or snake a drain instead of using a chemical drain cleaner.
- **Reuse:** Some hazardous waste materials may be reusable. Motor oil, for example, doesn't really degrade with use, and you can use it for other lubrication purposes instead of throwing it away. It's important to reuse wisely and know whether what you're doing is safe first.
- **Recycle:** Depending on where you live, you may be able to take certain hazardous waste materials to a treatment plant to be recycled. You can find out more about this by performing an internet search for recycling facilities in your area and inquiring about hazardous waste.
- **Donate Usable Leftovers:** Some hazardous waste materials, like caustic cleaners or lawn fertilizers, may be useful for others. Charities that work on home building or remodeling, including Habitat for Humanity, may be happy to take these items off your hands. Don't try to donate anything that's expired or otherwise unusable.
- **Do Not Mix Hazardous Materials:** It's essential to avoid combining hazardous waste materials when you gather them for disposal. Some hazardous materials can combine to create even more dangerous substances.
- Look Into Mail-In Options: Some waste disposal companies may offer mail-in programs accessible to anyone in the USA. This is a good option if you have certain potentially hazardous things to throw away, like compact fluorescent lightbulbs, but there aren't any local resources for safe disposal. Major national waste disposal companies like Waste Management may offer these services.

Medical Waste

All medical waste should be removed by a contracted medical waste disposal company and bagged in accordance with the species, condition, and medical waste company's policies. Proper PPE must be worn when handling medical waste.

Carcass Disposal

Deceased animals must be disposed of in ordinance with federal, state, and local authorities and should be approved by the Head of Animal Husbandry in advance. In most cases, the entity performing the necropsy will

be responsible for disposing of the carcass and other tissues and samples in accordance with all applicable regulations pertaining to the location of the facility, species of animal, and cause of death.

Veterinarian Procedures

Veterinary, medical, and animal care technicians may be consulted to assist with any animals' needs. The veterinary staff and Head of Animal Husbandry will investigate, research, and reach out to any specialist needed when and if a need arises. The goal will be to provide the animal with the best welfare possible and to maintain or enhance the animal's quality of life.

Hand Washing

All staff are required to wash their hands regularly when working with animals, after utilizing bathrooms, eating, sneezing, coughing, and touching dirty or contaminated surfaces. Staff are encouraged to utilize common sense and best practices to reduce disease transmission and maintain the health and safety of people and animals. Staff should wash their hands, or at least sanitize their hands, both before and after working with a species to limit transfer of pathogens to another species or staff member.

Photo IDs

All animals will have a photo taken when they arrive, and this photo will be updated throughout their life and if any changes occur in their appearance. These photos will be uploaded to in the animals record and sent to all appropriate parties.

Dire Wolf Disease Prevention

All animals will receive a periodic physical exam, monthly visual exams with BCS, formal welfare assessments done twice annually, and full physical exams will be done opportunistically if an animal is immobilized for any reason.

- Vaccines: Rabies and DHLPP puppy series and adult booster will be provided for each Colossal dire wolf. Puppy series will start when determined based on the health and wellbeing of the pups.
- **Parasite prevention:** Heartworm prevention will be given once a month and the type and dose of preventative will be determined veterinarians. The medication, dosing, and administration of the heartworm medication will be scheduled and recorded in the animal's record. Currently, the medication recommended is Simparico Trio.
- **Supplements:** Dire wolves fed a high-quality diet shouldn't need any additional supplements. If an individual needs supplements, the supplements will be determined by the Head of Animal Husbandry and the veterinarian.
- Fecal Test Schedule: March, June, September, and December
- Annual Preventative Testing: Annual blood collection and testing is based on a variety of factors, including testing brand, method, and life stage.
- Neonate Exams: To help ensure that exams will not cause maternal neglect or put staff at risk, neonate exams are done when the Head of Animal Husbandry and veterinarians determine it is safe to do so.
- **Training:** Animals will be trained to enter a species-specific restraint device to assist with voluntarily participating in medical procedures such as injections, blood draws, x-rays, and hoof trimming.
Occupational Health and Safety

Colossal takes safety seriously and does its best to encourage and provide safety measures for all staff. All required training is provided for all employees.

Occupational Health and Safety Overview

- All staff will be shown how to safely operate any vehicles/machinery needed for their routine. If you do not know how to operate something, do not use it until properly trained.
- All staff will be appropriately trained and prepared to respond to emergency situations through drills happening no less than once a year.
- All staff must use safety tools, procedures, and good judgment to maintain the health and safety of all animals and persons. See pepper spray section below and Lock Out Tag Out.
- All staff should utilize radios as the only source of communication when working around Colossal dire wolves. No cell phone communication, including texts or phone calls, should be made when working around the dire wolves.
- Personnel should assess all tools and equipment daily and throw out any that are not in good working order.
- Do not take shortcuts. Always follow the rules and use tools and equipment the way they are meant to be used.
- Staff must wear appropriate uniforms and PPE when working with animals.

First Aid and CPR

Each building, defender, quad, and all three jeeps are equipped with First Aid kits. All vehicles are equipped with trauma kits and AEDs (Automated External Defibrillator). All injuries must be reported to your supervisor.

Any staff member that is certified as a First Aid Responder, Mental Health First Aid Responder, and/or CPR certified will be listed for staff to contact in case of an emergency.

If you are injured, notify your supervisor. Injuries at animal facilities can have additional emergencies and a person knowledgeable in the area and routine will need to help assess, possibly secure animals, or identify substances that the injured party was working with.

All accidents, injuries, and near misses require an Accident, Incident, and Injury Report to be filled out and turned into your supervisor. See also any Facility Accident/Incident Report Forms and SOPs.

For life-threatening injuries, call 911.

An emergency contact list should be in each animal facility, the employee office, and break room. Give a precise address and building number, so the first responders can go to the closest entrance. The personnel monitoring the phones can guide EMS if required.

List personnel monitoring phone contact information here:

For non-life-threatening emergencies:

- Call supervisor
 - The supervisor on property will determine the type of care needed and contact 911 or a certified first aid responder on property.
 - They will contact the Head of Animal Husbandry to advise of the situation as soon as the situation is assessed and stable.

Urgent Care Center

If the injury is not life threatening or requires care that does not require equipment or medicine found only at the emergency room, the injured party should be taken to urgent care.

List locations of nearby urgent care centers:

List locations of nearby hospitals for emergency care:

Hand Washing

Hand washing is a simple and effective way to help prevent the spread of pathogens and disease. Colossal recommends that all staff at all facilities wash their hands following CDC guidelines. The CDC recommends washing your hands with antimicrobial soap and water often and during occasions where you are more likely to spread germs to other humans and in some cases animals.

- Before, during, and after preparing food
- Before and after eating food
- Before and after caring for someone at home who is sick
- Before and after treating a cut or wound
- Before and after using the toilet
- After <u>changing diapers</u> or cleaning up a person who has used the toilet
- After blowing your nose, coughing, or sneezing
- After touching an animal, animal feed, or animal waste
- After handling pet food or pet treats
- After touching garbage

Five steps to washing your hands every time, per the CDC:

- 1. Wet your hands with clean, running water (warm or cold), turn off the tap, and apply soap.
- 2. Lather your hands by rubbing them together with the soap. Lather the backs of your hands, between your fingers, and under your nails.
- **3.** Scrub your hands for at least 20 seconds. Need a timer? Hum the "Happy Birthday" song from beginning to end twice.
- 4. Rinse your hands well under clean, running water.
- 5. **Dry** your hands using a clean towel or an air dryer.

If soap and water are not available, you can use an alcohol sanitizer with at least 60% alcohol, and once you get to an area with soap and water, wash your hands. Make sure to cover all surfaces of your hands and rub your hands together until they are dry to correctly use the hand sanitizer.

Keep in mind:

- Hand sanitizers don't get rid of all germs.
- Hand sanitizers are not as effective if hands are visibly dirty or greasy.
- Hand sanitizers may not remove harmful chemicals from hands.

Definitions

Vector: any living agent that carries an infected pathogen and transmits that infectious pathogen to another non-infected living organism. (Examples: mosquitoes can carry the malaria parasite and infect humans with the malaria parasite. Other common vectors are ticks, raccoons, and rats.)

Fomite: an inanimate object that when contaminated with or exposed to an infectious agent can transmit the infectious agent to a new host. (Examples: animal bedding, nails, needles)

Reservoir: refers to a long-term host of a pathogen of an infectious disease, often the host of the pathogen does not get the disease carried by the pathogen. (Example: a reservoir for bubonic plague is rats.)

Disease Transmission

- **Direct Contact:** with an infected animal, saliva, blood, feces, urine, or bodily fluids. Contact can include petting and/or touching animals, bites, and scratches.
- Indirect Contact: contact with object surfaces or places animals live or have touched that have become contaminated with germs. Areas can include barn stalls, food and water dishes, and animal enclosures.
- Vector-borne: Being bitten by a species that is a vector for that disease, usually a tick, mosquito but mammals can also carry vector-borne diseases.
- **Food-borne:** Eating or drinking something contaminated with feces or urine from an infected animal or person.
- Waterborne: Drinking water or coming in contact with water that has been contaminated with feces or urine from an infected animal or person.

Disease Agents

- Bacteria: examples: Salmonella, staph infection
- Virus: examples: Rabies, Influenza, Lyme disease and Rocky Mountain Spotted Fever
- **Parasite:** examples: malaria, tapeworms
- Fungus: examples: ringworm, valley fever

How to prevent the spread of infectious disease and reduce the risk of acquiring a Zoonotic disease:

- Wash your hands often and properly.
- Wearing personal protective equipment (PPE). The use of PPE will reduce your risk of exposure.
- Reduce exposure to insects. Use appropriate bug spray, wear appropriate clothing, check yourself for ticks, and remove them quickly if you have been working outside.
- Avoid direct contact with animals: Only staff trained to handle animals should ever come in contact with wild or managed animals on property. If you are in an area with an animal and not trained to handle that taxa or species, don't interact with that animal without a trained staff member present.
- Make sure your required vaccines stay current, and if needed have your titer check.

Personal Protective Equipment

The purpose of personal protective equipment (PPE) is to protect the health and safety of the animals in our care, protect the health and safety of the people caring for the animals, and to protect our coworkers, friends, and families.

Handwashing is the most important thing a person can do to prevent the spread of pathogens, organisms, or contaminants to people or animals. Remember to wash your hands regularly and often following CDC guidelines.

It is your responsibility to utilize and adhere to personal protective equipment to protect yourself and the animals in your care. Protective clothing and equipment may include:

- Coveralls
- Boots
- Shoe covers
- Rubber gloves
- Vinyl gloves
- Disposable gowns
- Tyvek suits
- Face masks
- Face shields
- Goggles
- Ear protection
- Welding helmet, coat and gloves
- Respirators
- Sunscreen
- Bug spray
- Umbrellas and pop-up tents for sun protection
- Hand sanitizer
- Reflective safety vest
- Chainsaw chaps
- Assigned radio or cell phone
- Animal handling gloves with bite protection such as kevlar or reinforced leather

Each area should list where PPE is located at their building and what PPE is in each location.

PPE must not be worn out of a specific work area. Do not wear PPE out of that animal's area. Do not wear PPE in non-animal areas such as break rooms, bathrooms, meeting spaces. Take off your gloves if you are handling shared equipment that would not normally be contaminated (e.g., a computer, or phone).

It is required that staff have a dedicated pair of closed toe shoes that they wear only at work. They should change into their work shoes when they arrive and change out of them before leaving the grounds. Staff should change from street clothes to their uniforms at work and should change back out of their uniforms before leaving for the day. Shoes and clothing can transport pathogens to your home environment, and vice versa.

Staff should disclose to their supervisor of any pets they have at home, that way protective measures can be taken, if necessary, to protect both Colossal animals and the staff's personal pets. Staff may be required to show proof of their personal pets' current vaccination records to prevent bringing pathogens to the Colossal dire wolves, especially more vulnerable members of the pack, including pregnant dogs, pups, and medically compromised or geriatric individuals.

When caring for the Colossal dire wolves the following PPE must be worn:

- All shoes or boots must have closed toes.
- All staff must wear disposable gloves when preparing and handling meat. Because gloves may trap water against the skin, hands must be washed immediately after gloves are removed.
- Staff will utilize shoe in/shoe out to help reduce pathogens exposure to the animals and themselves.
- It is recommended staff wear hats and sunscreen if working in direct sunlight.

Pepper Spray

All facilities with Colossal dire wolves over 45 pounds will be equipped with pepper spray for use in the case of emergency such as an animal escape. Pepper spray is to only be used on animals which are attacking or appear likely to attack humans.

***Do not seek out encounters with dangerous animals. This product is a deterrent which may protect users in some unexpected confrontations but may not be effective in all situations or prevent all injuries.

Staff are not to use pepper spray on people or objects (there are no deterrent effects in doing so).

All staff should carry pepper spray when working around Colossal dire wolves over 45 pounds. Staff will be trained on the use of pepper spray. New trainees will be shown how to use pepper spray on their first day by senior animal care staff. Management will conduct annual training/refresher training as needed.

- Keep the safety clip on until you need to use the product.
- Place your forefinger through the hole in the handle with your thumb on the safety clip curl.
- With your thumb, pull the safety clip straight back and off.
- Depress actuator tab for bursts of spray.
- Aim at the face and eyes of animals.
- Depress actuator tab for 1-2 seconds to create a barrier of spray between you and the animal.
- Stop to evaluate the impact of wind and other factors and adjust your aim if needed before spraying again.
- Once safe, replace the safety clip by pushing firmly with your thumb until an audible "snap" is heard. Check to make sure safety is completely in place. No gap should be visible between actuator handle and safety clip.

Individual cans of pepper spray are labeled and will be checked yearly by senior animal care staff.

Pepper Spray Dangers

Pepper spray may cause irreversible eye damage if sprayed in the eye at close range. Contact through touching or rubbing eyes may result in substantial but temporary eye injury, strongly irritating mucus membranes and skin. Do not get in eyes, on skin, or on clothing. Wash thoroughly with soap and water after handling. Remove contaminated clothing and wash before reuse. It is best practice to keep MSDS sheets near where chemicals are stored, and have staff read them during their training in the areas.

First Aid Tips

- If in eyes: Hold eye open and rinse slowly and gently with water for 15-20 minutes. Remove contact lenses after the first 5 minutes, then continue rinsing the eye. Call the poison control center or doctor for treatment advice. Have the product container with you when calling or going for treatment. For emergency medical treatment information, call 1-800-535-5053.
- If on skin or clothing: Take off contaminated clothing. Rinse skin immediately with plenty of water for 15-20 minutes. Call the poison control center or doctor immediately for treatment advice.
- Physical or chemical hazards: Pepper spray is extremely flammable as contents are under pressure. Users should keep the canister away from fires, sparks, and heated surfaces. Do not puncture or

incinerate the container. Exposure to temperatures above 130° F may cause bursting, so pepper spray should not be left in a vehicle.

Considerations for pepper spray:

- This product has a range of up to 30ft (9 meters)
- The canister empties in approximately 7 seconds

For more information, refer to the USGS Bear Spray Training PowerPoint: <u>https://www.umt.edu/coop-unit/resources/employeeresources/usgs-bear-spray-training.pptx</u>



Accident and Incident Report Form Jan 2025

Accident, Incident, and Injury Report Form

Date: Time: Location:

Injury 🗆

Damage Only □

Near Miss/ Unsafe Condition

Details of injured person or main person involved in the damage or near miss: Full Name: Phone Number: Home address:

Employee: Employees Title: Contractors Company: Guest:

Contractor:

Describe how the injury, damage, accident or incident occurred.

Describe the type of injury, damage, accident or incident (example: deep laceration on hand, lock left unlocked, fence post bent).

How could the injury, damage, accident or incident have been prevented?

List of PPE personal protective equipment being used:

List machinery or equipment that was being used:

Name and contact details of all witnesses:

Name and Signature of person filling out the report:

Dire Wolf Transport Plan

The following procedures will be followed for the transport of dire wolves between animal facilities. Dire wolves may be transported for a variety of reasons including advanced medical care, relocation to a new facility, etc.

Animal Transport Personnel and Responsibilities

The planning and approval of the animal transport plan is the ultimate responsibility of the Head of Animal Husbandry. In this case, it has been determined to be appropriate to contract with a proven animal transport professional (someone who is experienced and licensed for animal transports) to oversee the transport details and the transport itself. In addition to the animal transport professional, a designated member of the animal husbandry team will also accompany the transport. This will include at least one of the following individuals: the Head of Animal Husbandry, the Chief Animal Officer, and/or the Manager of Animal Husbandry. Other members of the team and other designees may also accompany the transport upon approval by the Head of Animal Husbandry and the Chief Animal Officer.

Prior to Transport

The responsible parties at the sending facilities will ensure that the dire wolf/dire wolves are in good condition and determined to be healthy enough to travel safely. Dire wolves will have been properly fed, watered, and given the opportunity to urinate and defecate prior to being loaded into the transfer crate.

Animal Transport Vehicle and Logistics

The vehicle used will be large enough to accommodate at least the driver and one animal care technician, along with the dire wolf/dire wolves and can maintain a comfortable climate in the cargo area where the dire wolves will travel (cargo van or similar).

The appropriate capture equipment will be in the vehicle that transports the animals.

Any personnel beyond the driver and primary animal caregiver traveling in the animal transport vehicle will need to be safely seated while still providing ample room for the dire wolf crates. All other personnel will travel in chase vehicles.

The dire wolf/dire wolves will each travel in an appropriately sized container that complies with IATA Live Animal Regulations (refer to IATA Container Requirements, Container Requirements 82 document). This will be an aluminum exotic carnivore crate or similar, designed specifically to meet these standards. Crates will be properly secured to not move during travel or in the case of sudden stop. Crates will be positioned so that the main crate door faces the back or side of the vehicle. The crates will be labeled with Live Animal decals. Blankets and sheets will be on hand to allow crates to be covered in the case that the dire wolf/dire wolves appear nervous to help calm them by limiting sights from their surroundings, or if climate control were to fail, to keep dire wolves warm in case of colder outside temperatures. A water container will be affixed to the front of the crate. Water will be made available for the dire wolves in the crates. If more water needs to be added, this will be done easily without opening the crate. Food will also be added from outside the crate without opening the door if the duration of travel unexpectedly requires feeding.

Items to be included for transport:

- One crate per juvenile/adult canid, with a water vessel attached
- One sheet and one blanket per crate
- Supplemental water to fill and refill water vessels for animals for the duration of the trip
- Supplemental food in the event of unforeseen delay

- Towels in the event that a clean-up of the crate or van is needed
- Y-poles
- Catch pole with quick release
- Animal handling gloves with bite protection such as Kevlar or reinforced leather
- Capture net
- Ratchet straps
- Tool kit including, but not limited to, drill, screws, zip ties, duct tape (will include any other materials determined to be appropriate to address any concerns that may arise)
- The responsible veterinarian will be consulted regarding the chemical immobilization kit for recovery should an escape occur

In Case of Emergency

In the event of a vehicle issue or accident, animal injury or illness, or any other issue during transport, please refer to the document titled <u>USDA Animal Transport Contingency Plan</u> which also accompanies the transfer in the vehicle in hard copy form.

If an animal gets loose and is out of the immediate control of the transport team, local authorities will be contacted and briefed on the situation. All efforts will be made to partner with local authorities on recapture efforts. Additionally, team members will reach out to local zoos and wildlife partners for assistance with recapture efforts.

Important Contacts Numbers:

To be filled out with the specific contact information before each specific transport, based on participants and their roles in the transport.

Animal Transport Professional:

Head of Animal Husbandry:

Chief Animal Officer:

Manager of Animal Husbandry:

Clinical Site Veterinarian:

IATA Wolf Transport Requirements

The following information is an excerpt from the International Air Transport Association's Live Animals Regulations (LAR) Edition 50. These digital guidelines, purchased by Colossal, help ensure live animal shipments are handled and transported in the most expedient and humane manner and that animals arrive at their destination in good health. Facilities should always refer to IATA requirements to ensure they are following the current live animal regulations.

IATA (2023). Live Animals Regulations (LAR) Edition 50. IATA.

https://digitalpublications.iata.org/books/LAR-6105-50

https://digitalpublications.iata.org/reader/books/LAR-6105-50/epubcfi/6/588%5B%3Bvnd.vst.idref%3DLAR-8-ContReq-82%5D!/4/4%5BLAR-8-ContReq-82%5D/20/4/1:64%5B%20no%2Ct.%5D

Container Requirement 82

The illustrations shown in this Container Requirement are examples only. Containers that conform to the principle of written standards for the species but look slightly different will still be considered compliant with the IATA minimum standards.

Applicable to: Aardwolf Andean mountain cat Asiatic wild dog Badger species Bobcat Bush dog Caracal Coyote Dhole Dingo Dog, bush wild Dog, hunting wild Fox species Hyena species

Jackal

Jaguarundi

Lynx species

Maned wolf

Ocelot

Otter species (except giant otter, see CR80)

Panda (lesser or red)

Serval

Wild cat species (small)

Wolf

Wolverine

STATE VARIATIONS: GBG-01 GBG-02 GBG-03 GBG-04 GBG-05 HKG-01 SAG-02 USG-08 USG

OPERATOR VARIATIONS: AF-01 AC-05 BA-04 CX-05 CX-07 IB-02 GF-07 GF-11 KL-02 MK-01 MS-01 QF-01 SV-01

1.CONTAINER CONSTRUCTION

Principles of Design

The following principles of design must be met in addition to the General Container Requirements outlined at the beginning of this chapter.

Materials

 \triangle Wood, metal, synthetic materials, welded wire mesh.

Dimension

The height of the container must allow the animal to stand in a natural position with its head extended and the width must permit it to turn around and lie down comfortably. The actual measurements will vary with the species involved.

Frame

The frame must be made from solid wood or metal parts bolted or screwed together. It must be constructed so that it cannot be damaged from continual biting or scratching at the corners. If the total weight of the container plus animal exceeds 60 kg (132 lb.) metal bracing must be added to the frame.

Sides

 \triangle The sides and door must be made of metal or solid wood. The front of the container must be constructed of welded wire mesh. The mesh must have a diameter that will prevent the animal protruding its nose or paws to the outside. The whole front must be covered by a sliding shutter that is offset from the welded wire mesh by approximately 5 cm (2 in) and which can be raised and lowered to permit feeding and watering. It must have

two observation holes of at least 10 cm (4 in) in the upper part and ventilation holes, with a minimum diameter of 2.5 cm (1 in), spread over the remainder of the surface in order to give good ventilation but at the same time leave the animal in semi-darkness.

Floor

The floor must be slatted, over a leak-proof droppings tray or, if slatted floor is not required for that species, it must be leak-proof and covered by sufficient absorbent material in order to prevent any excrete escaping.

Roof

Must be solid wood or metal.

Doors

 \triangle A sliding door must be provided for all non-domestic species, it can be made from the welded wire mesh ventilation front if required. It must have a secure means of fastening so that it cannot be opened accidentally.

Ventilation

The main ventilation front must be supplemented by meshed openings along the upper part of the container walls and/or holes with a minimum diameter of 2.5 cm (1 in) spread over the top third of the sides and the whole of the back. These holes must be spaced both horizontally and vertically at intervals of approximately 10 cm (4 in) center to center. At least one-third of the total ventilation openings should be on both the lower and upper half of each ventilated wall.

The total ventilated area must be at least 16% of the total area of the surface of all four sides. More ventilation and the use of larger meshed openings is permitted but the animal must not be able to protrude its nose or paws to the outside from any opening.

If the mesh is fixed to the interior of the container all sharp edges must be protected.

Spacer Bars/Handles

Must be made to a depth of 2.5 cm (1 in), must be present on the sides of the container as shown in the illustration.

Special Requirements

Hyena, wolves, badger, otter wolverine and wild dogs must have the container completely lined with sheet iron or other hard metal sheeting with through ventilation holes cut into it.

 \triangle Palletized shipments must have the containers made entirely of welded wire mesh of a suitable dimension that no part of the animal can protrude in order to ensure good ventilation.

Sea otters require a slatted floor, within the leak-proof container, to ensure that waste does not remain in contact with the animals. Where used for sea otters, Rigid Plastic Pet Containers should be modified with a slatted floor.

Forklift Spacers

Must be provided if the total weight of the container plus the animal exceeds 60 kg (132 lb.).

Rigid Plastic Containers

(see Container Requirement 1)

Some of the less destructive of these species can be transported individually in modified rigid plastic pet containers. Rigid plastic containers are not suitable for African wild dogs.

The Rigid Plastic Container must meet all requirements in CR1 of the current LAR. In addition, the following modifications must be made:

•all ventilation openings must be covered with welded wire mesh.

•the door must have secure fastenings at the top and the bottom.

•a curtain, which can be raised and lowered and does not impede ventilation, must be fixed over the door to reduce light inside the container.

2.PREPARATIONS BEFORE DISPATCH (see Chapters 5 and 10)

No special requirements.

3.FEEDING AND WATERING GUIDE

Food and water containers must be provided with a means of access from the outside.

Animals do not normally require additional feeding or watering during 24 hours following the time of dispatch.

If feeding is required due to an unforeseen delay, canned dog or cat food must be provided but care must be taken not to overfeed.

For sea otters, regular feeding is required. Provision for refrigerated seafood items must be made for any transport. Sea otters also require continuous access to fresh water ice for cooling and consumption throughout the transport.

4.GENERAL CARE AND LOADING (see Chapters 5 and 10)

Animals in quarantine must be segregated from those which are not.

Hand-reared young may be loaded in the same container as long as they are used to cohabiting.

Cargo compartment temperature and ventilation requirements should be discussed with the operator prior to the transport.

Transport at lower cabin pressure altitudes is preferable for sea otters. Special arrangements should be made with the operator prior to the transport.

IATA Pet Container Requirements



CONTAINER REQUIREMENT 1

The illustrations shown in this Container Requirement are examples only. Containers that conform to the principle of written standards for the species but look slightly different will still be considered compliant with IATA minimum standards.

IATA activities are limited to the development of standards for the acceptance, packing and handling of live animals for transportation by air. IATA does not certify, approve, endorse, or sell any particular pet container manufacturer, brand, make, or model. Equally so, IATA does not offer, solicit, endorse, or approve any particular pet or puppy transport or relocation services, regardless of whether these be offered via email or the internet. Readers should pay attention to fraudulent offerings that claim the opposite.

Applicable to:

Cats (domestic)

Dogs (domestic)

STATE VARIATIONS: CHG-01/03/04, GBG-01/02/03/04/05, HKG-01, NZG-01, SAG-02, ZWG-02/03/04, EUR-01, USG-Variations

See exceptions AUG-01 and NZG-01 in Chapter 2; Some state regulations require that the container must be sealed during transportation.

OPERATOR VARIATIONS: AC-03/04/07, AF-01, CX-05/06/07, EI-01, EK-05/07/08/12, GF-05/06/10/11, KL-01/02/08/09, LH-05/08/10, LX-05/07, MK-02, MS-01, OK-01/06, PR-01/04/05/06/07/08, TG-01, QF-02/03/04/07, UA-07/08/09

AC-01 and QF-01 in Chapter 3; Certain operators will not carry wooden containers.

Note:

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For carriage of domestic pets in passenger cabins as accompanied baggage see Chapters 2 and 3.

For animals travelling in the aircraft cabin the travelling non-rigid container must:

- Meet these container requirements except that the container construction does not have to be rigid and spacer bars are not required.
- Allow the animal contained in the container to have enough space to turn about normally while standing, to stand and sit erect, and to lie in a natural position.
- The ventilated area must be at least 16% of the total surface of the four sides of the containers.
- The container must be clearly marked with a Live Animal label and the label must not block any of the ventilation openings.
- Meet these container requirements except that the container does not need to have water and food containers.
- The requirement for one end of the container to be welded wire mesh does not apply to soft side/cloth bags.

The check-in staff is responsible for ensuring the container meets the IATA requirements. Should the container not meet the minimum standards the animal must not be allowed to travel.

1. CONTAINER CONSTRUCTION

Principles of Design

The following principles of design must be met in addition to the General Container Requirements outlined at the beginning of this chapter.

The interior of the container must be smooth with no protrusions that the animal can bite or scratch to cause damage to the integrity of the container in any way.

All openings must be nose and paw proof to avoid injury to the animal and handlers.

Openings must be a maximum of 25 mm x 25 mm (1 in x 1 in) for dogs and 19 mm x 19 mm ($\frac{3}{4}$ in x $\frac{3}{4}$ in) for cats. The openings may have to be smaller in order to be nose and paw proof.

If a container has wheels, they must be removed or rendered inoperable.

Materials

Fibreglass, metal, rigid plastics, welded wire mesh, solid wood or plywood.

All wood used for the construction of containers for international transport must comply with IPPC standards.

For all containers the welded wire mesh must be 2.5 mm or thicker for dogs and 2.0 mm or thicker for cats.

Containers made entirely of welded mesh or any other type of wire mesh are not suitable must not be used for air transport.



Plastic: Rigid plastic containers \checkmark are suitable for most breeds of dogs but their acceptability is at the discretion of the operator. Some rigid plastic containers may not be suitable for large dogs, or dogs that are aggressive.

Example of a rigid plastic container and an example of a wooden container:

EDITION 49, JANUARY 2023



Figure

TYPICAL RIGID PLASTIC CONTAINER

Locking pins must engage the container beyond the extrusions by at least 1.6 cm (% in)





Back view

Size (Mandatory performance requirements when selecting a shipping container)

Each animal in the container must have enough space to stand, to sit erect, to lie in a natural position and to turn about normally while standing.

Guidance on Calculating Minimum Container Dimensions

To assist in determining the approximate size of the traveling container, the following formula may be useful. However, it may need to be adjusted to meet the mandatory performance requirements listed previously. When calculating the minimum internal height of the container, the height of bedding should be added to the height of the animal.

Animal measurements:

A = length of animal from tip of nose to base/root of tail.

B = height from ground to elbow joint.

C = width across shoulders or widest point (whichever is the greater).

D = height of animal in natural standing position from top of the head or the ear tip to the floor (whichever is higher).

Note:

Measurements A, B, C and D for determining the container dimensions must relate to the largest animal.



The calculated dimensions are internal container dimensions.

Minimum internal container dimensions for a single animal:

Container length = A + 1/2 B

Container width = C x 2

Container height E = D + bedding

Snub-nosed breeds require 10% larger container.

Note:

Container width calculation for multiple animals:

- Two animals: C x 3
- Three animals: C x 4

The height and length are determined the same as for a single animal.

Crating animals together:

Weaned puppies or kittens may travel well together in the same primary enclosure. When crating puppies or kittens together in the same container/primary enclosure they must be from the same litter, not older than six months, weigh no more than 14 kg each and no more than three per container. Certain national regulations require cats or dogs to be crated individually unless the consignment is a litter over 8 weeks and travelling with the mother.

A maximum of two adult animals of comparable size up to 14 kg each, that are compatible in size and used to cohabiting, may be shipped in the same container/primary enclosure. Animals over 14 kg must be crated individually.

Sides

Side walls must be solid with sufficient ventilation as prescribed.

Plastic containers: Where containers are assembled from a top and bottom part, these must be securely bolted together. Plastic clips must not be the only fasteners holding the top and bottom part together, the use of metal nuts and bolts in every hole is recommended.

Wooden containers: For containers made of wood, or plywood, the minimum thickness of the sides is 12 mm (1/2 in) for dogs and 6 mm (1/4 in) for cats.

Floor

The floor must be solid and leak-proof.

Roof

The roof must be solid, but ventilation holes and welded wire mesh are allowed over the whole surface provided that they do not reduce the integrity of the container and the strength of the roof itself.

Wooden containers: For containers made of wood or plywood, the minimum thickness of the roof must be $12 \text{ mm} (\frac{1}{2} \text{ in})$ for both dogs and cats.

Door

The door must be constructed of plastic, wood, plywood, welded or cast metal of sufficient thickness so as to preclude the animal from bending or distorting the door.

The door must form the whole of one end of the container. It can be either sliding or hinged.

For all containers using doors with locking pins, the pins must engage the container by at least 1.6 cm (5/8 in). The shipper must ensure that all hardware and fasteners are in place and serviceable.

Plastic containers: Doors made of plastic are permitted, provided that hinges and locking pins are made of metal of sufficient thickness.

When rigid plastic containers are used the door should be further secured by application of additional removable fasteners such as cable ties in the four corners of the door frame. Care must be taken when selecting the fasteners to ensure these will not cause injury to the animal or to the handling personnel.

There must be an adequate means of fastening and sealing for containers travelling to countries where sealing is required.



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Ventilation

The total ventilated area must be at least 16% of the total surface of the four sides. The provision of additional ventilation openings on the roof or sides of the container or larger ventilation openings covered in welded wire mesh in order to increase the ventilation are permitted.

Ventilation must be provided on all 4 sides of the container. Ventilation openings on the sides and the back must be placed over the upper two thirds of the container.

△ The whole of one end of the container (which can be the door) must be ventilated.

The welded wire mesh insert covering ventilation openings must be securely fixed to the container so that the animal cannot dislodge it.

△ All openings must be nose and paw-proof, in the case of cats and small dogs these may have to be covered with a second layer of welded wire mesh. Where a second layer of welded wire mesh is required to make the container nose and paw proof both layers of mesh must meet the minimum standards prescribed for the species. It is very important that no animal has any surface or edge at which it can gnaw or scratch.



Wood/Plywood Containers

Where ventilation is provided by welded wire mesh it must fully overlap the frame. It must be attached to the outside of the frame using staples at least 19 mm ($\frac{3}{10}$ in) long and at intervals of no more than 25 mm (1 in). In addition, for dog containers made of wood/plywood, a wood/plywood frame or strips of wood/plywood must be

placed over the edges of the welded wire mesh and affixed with screws. This compresses the welded wire mesh between the strips of wood/plywood and the outer surface of the container. The wood/plywood compression strips/frame must be at least 12 mm thick and of a placement to adequately secure the wire. Fasteners must not penetrate into the container.

Spacer Bars

Must be provided along both long sides of the container. A spacer bar must also be provided along the back of the container.

Spacer devices must be positioned to prevent other freight from blocking the ventilation.

Handles

Appropriate handles must be provided. Spacer bars where suitable may be considered as the handles. Handles must provide the handlers a means to move the container without a risk of being scratched or bitten by the animal.

Forklift Spacers

Must be provided for all containers where the total weight with the animal exceeds 60 kg (132 lb) and must be a minimum 5 cm (2 in) in height.

Bedding

Absorbent bedding that is suitable for the species must be provided.

SPF Containers

Specific Pathogen Free (SPF) dogs and cats must be transported in containers that conform to the requirements published in this Container Requirement. A "Laboratory Animals" label must be affixed to the container and "This Way Up" labels must be placed on at least two opposite sides. Filter containers for SPF consignments have special gauge air filters fixed in the ventilation apertures. Ventilation must be a minimum of 16% of the surface area of the four sides. Sufficient water must be provided for the journey. Food must be provided, if required, at the point of origin in order that the sealed container is not opened during transport. A viewing panel must be provided on SPF containers.

Labelling

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A green "Live Animals" label/tag or a red "Laboratory Animals" label/tag is mandatory on all live animal consignments. "This Way Up" labels/tags are also mandatory and must be placed on at least two opposite sides. The label or tag can be imprinted on the container.

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It is recommended to mark the animal's name on the container.

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2. PREPARATIONS BEFORE DISPATCH (see Chapter 5)

Tranquillization of dogs and cats is not recommended.

Sedation of animals, except under certain conditions and carried out under veterinary direction, is not recommended. Most, if not all, commonly used tranquillizing drugs have the effect of lowering the blood pressure, this also occurs naturally at high altitudes. The air pressure of an aircraft in flight is set at the equivalent of approximately 8,000 ft.

The combination of altitude and drugs is potentially fatal in the old, chronically sick or stressed animals. Calming by darkening the container and putting it in a quiet place when not in the aircraft, will calm most animals. If sedatives are used, the name of the drug, the time and route of administration must be clearly marked on the container and a copy of the record must be attached to the documents relating to that shipment. Any further medication administered must be recorded and accompany the shipment with the name of the sedative, time of administration and the route of administration.

Shipment of females in heat (oestrus) is not recommended.

Females with suckling young and unweaned animals must not be accepted for carriage.

Weaned puppies and kittens younger than eight weeks must not be shipped due to possible dehydration effects in air transportation.

Dogs and cats should only be shipped during the first two thirds of pregnancy. During the last one third of pregnancy the chance of spontaneous abortion or injury to the fetus increases. Significant debilitation or death of the mother can be initiated by the stress that may occur during transportation at this time in gestation.

Dogs and cats should not be shipped for seven days following giving birth.

It is recommended that the shipper removes collars/ vests/harnesses/clothing and electronic GPS trackers from animals prior to crating. If GPS trackers or any other recording equipment are used, they must fully comply with IATA Dangerous Goods Regulations and must be declared to the operator.

□ For pet animals, a familiar article in the container helps to placate the animal. It is good practice to mark the animal's name on the container.

3. FEEDING AND WATERING

A water container must be present and affixed within the shipping container with outside access for filling that does not require the opening of the shipping container allowing potential escape of the animal. Food containers must be present either within the container, if sealed, or attached to it. Food may be attached to the travelling container for use in case of delay. Note illustrations are examples only, containers or receptacles that are attached may look different provided they meet the standards described.

Note:

- Water containers must be open on the top surface allowing the animal unrestricted access and must be securely affixed to the inside of the shipping container so they can't be dislodged.
- Water bottles or similar dispensers that are attached either inside or outside the shipping container are not considered to be open containers and shall not be used to replace open containers.

Feed the animal a light meal at least two hours before dispatch, provide a short drink and exercise the animal immediately before crating.

Animals do not normally require additional feeding during 12 hours following the time of dispatch. Water must be provided if total journey time exceeds 12 hours. Care must be taken not to overfill the container.

If feeding is required due to an unforeseen delay, meat, biscuits or canned pet food must be provided but care must be taken not to overfeed. Animals must not fly within 2 hours after their meal.

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4. GENERAL CARE AND LOADING (see Chapters 5 and 10)

△ Containers of young animals of the same species may be loaded adjacent to each other. Containers with cats and containers with dogs must be kept apart, unless they are used to cohabiting. Care must be taken in loading different breeds of dogs to prevent snapping an disturbing one another and, in particular, where one animal is stronger than the other, subjecting the weaker of the two to fear.

Accompanied dogs and cats that are transported in the aircraft hold must comply with these Regulations.

Warning 1: Snub-nosed dogs must be stowed as far away as practical from other loads to ensure they have the largest amount of air space available in the hold.

Warning 2: If it is necessary to open the container for any reason, this must always be done in an enclosed area in order to prevent the animals from escaping.

Animals travelling under quarantine must be segregated from those which are not.

USDA Animal Transport Contingency Plan

In the event of an emergency while transporting animals between animal facilities, these procedures will be followed. This document will accompany all animal transports.

Vehicle Issues or Accident

In the event of minor vehicle issues (flat tire, minor accident, etc.), ensure staff and animals are in a safe location. If further assistance is needed, please contact the Head of Animal Husbandry and/or Manager of Animal Husbandry to help determine locations to take vehicles for maintenance needs and/or safe places to hold animals while the vehicle is repaired. Alternate transportation will be identified from a nearby vendor or from a nearby partner zoological facility if needed.

Animals may be relocated to another safe space if needed and should always be under direct supervision of a team member or the designated animal transport professional. Contact 911 if staff and/or animals are in jeopardy. In the event of an emergency where a vehicle crash has rendered team members unconscious, this SOP will be posted in each vehicle as reference for emergency responders.

Animal Injury or Severe Illness

If an animal becomes ill or is injured during transport, the senior most personnel not in the vehicle of the animal will be notified immediately if the Head of Animal Husbandry and/or Manager of Animal Husbandry are involved with the transport. If the Head of Animal Husbandry and/or Manager of Animal Husbandry are not involved in the transport, they should be notified immediately. If a severe injury/illness occurs, the transport may be rerouted to a nearby zoological or veterinary facility at the leadership team's discretion.

Emergency Phone List:

To be filled out with the specific contact information before each specific transport, based on participants and their roles in the transport.

Animal Transport Professional:

Head of Animal Husbandry:

Manager of Animal Husbandry:

Chief Animal Officer:

1ST CALL:

2ND CALL:

3RD CALL:

4TH CALL:

5TH CALL:

Staff Training, Safety, and Accountability

Colossal dire wolves will be managed in a protected contact situation. Dire wolves should be trained to shift into holding areas and crates for transport and medical procedures. Shared space will be allowed for domestic dogs, pups, and juveniles 45 pounds or less for management purposes per agreed upon procedures. If the non-domestic dog is a dire wolf over 45 pounds, staff must contact the Head of Animal Husbandry for permission before entering the space for any reason.

Training Directive

All staff and other people in and around Colossal dire wolves, the holding areas, and habitats will be appropriately trained to work with and around large carnivores/omnivores and specifically the Colossal dire wolves. No caregiver should work with or around the area without being trained and a supervisor approving and verifying that it's safe for the specific caregiver to do so. Colossal dire wolf-trained staff should be assisting the area and be responsible for and prepared to respond and care for the dire wolf in emergency situations.

Individuals involved in the care of Colossal animals must use safety tools, procedures, appropriate PPE, and good judgment to maintain the health and safety of all animals and persons. Dire wolf caregivers should follow the Lock Out Tag Out procedures and all guidelines set by the Head of Animal Husbandry.

Communication and Cell Phone Usage

All staff should use radios as their main source of communication when working with dire wolves (**no cell phone communication either through texts or phone calls should be made when working around dangerous animals**). If staff need to use the phone, they should make sure all dire wolves are secured correctly and step away to safely make the phone call or text message.

Staff should follow the cell phone and smartwatch policies, such as placing cell phones in the cell phone bin when shifting/working around the dire wolves, which are classified as dangerous animals. Staff working around dire wolves should be focused solely on what they are currently doing; cell phones and smart watches provide unexpected distractions from the task at hand.

Personnel Safety

At no time should any person, regardless of title, ever enter the same space as a Colossal dire wolf weighing greater than 45 pounds. They should never enter a space the dire wolf has access to, regardless of the location of the dire wolf. Facilities may require all animal care technicians working around dangerous carnivores to always carry pepper spray on them, wear the proper uniform, and adhere to the uniform policy.

Staff will all be trained and aware of safety procedures to minimize/eliminate risk of injuries and zoonotic disease. This will be the responsibility of the facilities management and supervisory staff. Records of training for Colossal animals will be provided to Head of Animal Husbandry. Staff and any person working with Colossal animals are required to report any animal bites, scratches, stings, etc. Any occurrences must be reported in an Incident Report submitted to appropriate management within 24 hours.

For further details please see:

- <u>USGS Bear Spray Training</u>
- Accident, Incident, and Injury Report
- <u>Protected and Free Contact</u>
- <u>Shifting Procedures and Precautions</u>
- <u>General Communication Guidelines</u>

Lock Out Tag Out

Lock Out Tag Out (LOTO) is a type of program to help ensure individuals are protected from unexpected outcomes when working with dangerous animals/equipment or in dangerous situations. A Lock Out Tag Out system must be utilized to mitigate the risks of working with dangerous animals.

Lock Out: The practice of using keys or a combination of security locks to prevent the unwanted opening/ closing of doors/gates or other equipment.

Tag Out: The practice of using tags in conjunction with locks to increase visibility and awareness of doors/ gates or other equipment, to prevent the doors/gates and equipment from being moved until the Tag is removed.

Examples of LOTO equipment:

- Locks
- Tags
- Chains
- Wedges
- Self-locking fasteners
- Other devices for isolating, securing, or blocking doors/gates or equipment from moving

Choosing Devices

When choosing devices to work with an area, enclosure, and/or facility, keep in mind:

- The device chosen for Lock Out should be used for controlling the movement of the door/gate, should only have one purpose, and serve no other function.
- The device should be able to handle the environment it's being used in for the entirety of the time it needs to be employed.
- Weather should not cause the Lock Out device to degrade/deteriorate. The environment should not cause the Tag or Lock Out device to corrode.
- Tags should be standardized by color, shape, and/or size.
- The Tag and Lock Out device chosen should be robust enough to prevent inadvertent removal without the use of excessive force or bolt cutters.
- The Lock Out Tag Out procedures should include surveys to locate and identify all isolating doors/gates and equipment, including but not limited to switches, valves, control handles/buttons to be locked out.

• Employees should be trained on all aspects of the LOTO SOPs and procedures which will include the type and magnitude of the energy/force of the doors/gates and equipment being controlled and stopped from moving.

Lock Out Steps

Personnel should be assigned to individual locks or tags to ensure the system can't be bypassed.

All LOTO devices and programs should be tested without compromising the security or safety of staff or animals.

LOTO devices should only be removed after servicing/maintenance is complete and people/equipment are out of the area.

Examples of LOTO Systems

- 2 animal care technicians, 2 different keys: all doors are locked with two separate keys that two different people are assigned and are required to lock/unlock any doors in dangerous animal areas. No door can be moved without both animal care technicians unlocking it. This gives a second set of eyes on all locks. No animal can be shifted and no person can enter or shift without the second person there to help ensure all parties are safe and secure at all times.
- A general lock that is keyed differently from the other locks that goes on in conjunction with a standard dangerous animal lock system: All dangerous animal staff have their own lock assigned to them with their own key, that no other dangerous animal care staff has. The animal care technician assigned to the area for that day places their lock on each door in a spot designed to be the second lock on that door. This ensures that no other person can inadvertently move doors without the assigned animal care technician being there to help ensure animals remain secure and are not sharing a space with a human or other animal they should not have access to. At the end of the day the animal care technician removes their assigned lock leaving the area secured with the general dangerous animal area locks.

Resources for the Lock Out Tag Out Program include:

OSHA: <u>https://www.osha.gov/sites/default/files/publications/osha3120.pdf</u> **Safety Culture:** <u>https://safetyculture.com/topics/lockout-tagout/</u>

Animal Escape

Once a Colossal dire wolf reaches a weight of 45 pounds, it will be treated as a large carnivore and considered dangerous. Any domestic dog will not fall into this category. No document can give direction on every specific situation or scenario, so this document serves as a guideline. The best way to handle animal escapes is to prevent them from happening. Staff should always be conscious and diligent of prevention. Staff should practice the following habits as well as endeavor to identify and communicate additional habits, fail safes, and systems to prevent animal escapes:

- Always remain calm.
- Avoid carelessness, inattention, and distractions especially when opening doors/gates and when entering/ exiting buildings and habitats and shifting animals.
- Always double check yourself and teammates. Physically make sure all doors and gates are properly secured.
- Be aware when you are called away from your normal routine. These are times when locks, doors, and gates are most likely to be forgotten, left open, or unsecured.
- Know where your animals are at all times and count them before you begin any routine or procedure. If animals' whereabouts are not visually seen, practice caution when entering an area to get a visual confirmation on the animal.
- If you are working in pairs/groups, check with/for your partner, watch out for their safety as well as your own.
- Learn to anticipate dangerous situations before they begin.
- All animal care staff should be familiar with the location of capture equipment.
- Prepare your day, routines, and procedures with a safety-first attitude.
- Each morning and evening, animal care staff will make rounds to count animals and inspect enclosures. This is part of the normal animal care daily routine.

Definitions and Roles

An animal escape is called when an animal is not in its designated enclosure or otherwise under the appropriate control of an authorized person or persons. The concerns in an animal escape shall be prioritized in the following order:

- 1) Ensuring community safety
- 2) Ensuring outside safety personnel safety
- 3) Ensuring staff safety
- 4) Ensuring animal safety
- 5) Recapture of the animal(s)

Primary containment is an animal in its own enclosure.

Secondary containment is an animal in basic confinement, but not enough to keep it from escaping into a public area.

Secure secondary containment is an animal confined in a space that is not its normal space but is adequate to keep it from escaping into a public area.

First Responder is the first person to be aware of the animal's escape. They will radio the other animal care staff and security of the animal escape and clearly provide who (both species and name), what and where. They will send a group text message to inform the rest of the farm staff of the escape and inform more senior personnel, as well as the firearms team, to be on standby.

Incident Commander: This person will remain the incident commander until the senior most animal personnel or staff member can take over.

• The incident commander will have contact and rapport with Colossal and other outside entities, in-depth knowledge of the facility, and knowledge of recapture techniques, staff, vehicles, and tools. They will work with and direct all responding staff. They will be the person directing and coordinating the recapture on the radio. They, along with Colossal, will make the decision if it becomes necessary to have the firearms team shoot to kill.

Recapture Coordinator is the most experienced animal personnel with escaped animal(s) but may be a senior member of the animal care staff that is familiar with the animal in question and the facilities or the first responder. This person may also be the Incident Commander. This person should know all the details of staff on-grounds at the time of the incident. In addition, this person will know locations of gates, doors, and fences in relation to the area of the escape/incident and should also know where all recapture equipment is located. They will utilize their knowledge of the specific animal/animals to help ensure the safe recapture of the animal, with the most positive outcome possible. They will communicate their knowledge to the IC. They will not be directing the staff responding, they will be assisting the IC.

Firearms Team: Staff members trained in gun safety that have trained and qualified to be proficient in shooting all the guns found on grounds and are trained to shoot to kill animals quickly and humanely. All firearms team members are experienced hunters that practice on range outside of hunting season. This team will always respond in the case of a Colossal dire wolf escape.

Response Team: Animal care staff members that are trained in recapture techniques for the species in question can and should include veterinarians or personnel trained in safely immobilizing the species in question. The staff involved will usually be the animal care technician for the area or animal care technicians trained in similar taxa, such as the large carnivore animal care technicians. This team may include facility or maintenance personnel as they will know how to utilize any heavy equipment that might be needed. The animal care manager, lead animal personnel, veterinarian, and/or veterinarian technician or other personnel instructed by the Incident Commander will get capture equipment, drugs, and immobilization equipment as needed. At this time, the veterinarian, veterinarian technician, animal care manager, or other trained personnel will dispense any chemical immobilization.

Security: Security should include a list of the security personnel and local police phone numbers.

Drills are conducted at a minimum of once a year. Drills may include but are not limited to tabletop discussions and in-the-field practice scenarios. All drills will be documented and stored in accordance with all accrediting organizations requirements. Drills will be conducted with darting equipment regularly to ensure a swift and accurate response to an emergency.

Animal Care Personnel will be familiar with where the capture equipment is kept on the farm (nets, catch pole, gloves, dart gun, etc.). They will also regularly practice the safe handling use of all capture equipment.

Animal Care Management includes the veterinarian, animal care manager and lead animal personnel. They are to conduct monthly checks to ensure all equipment is sound and ready for emergency use. Dart gun practice occurs monthly, usually when a veterinarian is on-site, however, it can happen without a veterinarian.

Veterinarian, Animal Care Manager, or Curator will notify police, local, and state authorities that a dangerous animal has escaped and alert them of the situation. They will also start communication with on-site staff through group text and radio communication. **Initial Response**

- STAY CALM. How you respond emotionally, mentally, and physically will affect the animal's behavior and the people around you. Do not do anything to aggravate or excite the animal. Stay safe and keep your distance; approaching or startling an animal may cause it to flee. Try not to lose visual contact with the animal, understanding that you must stay safe.
- The first person to be made aware of an animal escape will radio other animal care staff and security of the animal escape. Give clear and precise details of who, what, and where. This person is considered the Incident Commander in the situation until the senior most animal personnel or more senior personnel can take over.
 - Radio communication will be limited to just the animal recovery response. No other radio communication will be permitted.
- The first to respond will send out a group text message to inform the rest of the farm staff of the escape and inform more senior personnel as well as the firearms team to be on standby.
 - Phone communication will be limited to just the animal recovery response. No other phone communication will be permitted.
- Responding personnel should simultaneously be:
 - Checking the perimeter gates and making sure they are closed and secure to prevent any animal from escaping the immediate area.
 - Heading to the treatment room to obtain the Immobilization Kit and dart gun and head to the location of the escaped animal in a covered vehicle.
 - Try to get as close as possible without evoking flight or fight response from the animal.
 - Take your time in getting a good shot.
 - Have the firearms team on standby in the event of any escape if lethal actions are needed. Nonfirearms team members or security should not shoot a Colossal dire wolf without the IC giving the order to shoot. Reasons for lethal action are escaping perimeter, or if personnel or community safety is compromised. The gun safe is located at the HQ building. On-site security will be armed with variable calibers 24/7.
 - Based upon the demeanor and body language of the escaped animal, the on-site security officer has the authority to determine if lethal or non-lethal intervention is required.
 - The veterinarian, animal care manager or curator will notify law enforcement and local/state game authorities if needed
 - Staff that respond should be delegated these tasks by the IC or notify the IC with what you're doing to take action to recapture the animal.
- If the dire wolf is a threat to the community or staff safety and cannot be quickly immobilized or returned to the facility, the police must be notified immediately. At this point the animal would be considered shoot to kill and if the firearms team has a shot, they should take it.

- Firearms team should verify that no person is behind the animal in case the shot doesn't hit the intended target.
- Firearms teams should ensure that they are not shooting at each other.
- They should make the IC aware of their location when they arrive on scene, so IC can help to prevent friendly fire or accidentally pushing the animal into other responders.
- Once the animal is safely recovered, an all-clear should be given to resume normal operations.
- There will be an immediate staff briefing on the escape, the procedures, and anything else pertinent to this subject. After the event, there will be a meeting on how to prevent the situation.

The initial call may be escalated to an additional threat (e.g., Animal Attack or Threat of Attack, threat of breaching secondary containment at any time), therefore the appropriate firearms will be brought to any animal escape involving the Colossal dire wolf.

If the person calling the escape does not have eyes on the animal, they should make that clear and the first staff member to have eyes on the animal will be the Incident Commander until a more senior or experienced staff member with the species involved arrives on the scene. In some cases, the Incident Commander may not have eyes on the animal and may have to rely on another staff member to relay information about the animal and situation.

The IC is responsible for ensuring staff safety by knowing where all staff, where the animal is, and where the animal is heading. The IC will immediately respond and set up a command center to safely coordinate all units responding to the escape. This may be a designated meeting spot or a place where they can safely coordinate all the responding units.

If it is safe to do so and will not scare the animal into a public area or further away, try to get some level of containment on the animal. This could be as simple as shutting the door to the building or back area. Do not shut any door that will trap anyone in with the animal.

Responding personnel should go to the area advised by the Incident Commander or the designated meeting point. Responding and approaching any area including preset areas of refuge should be done with extreme caution to maintain human safety and to not decrease chances of quick recapture. Do not proceed to any area without a designated meeting point. If the IC has not designated a meeting place within the first 1-2 minutes responding personnel should ask for one. Do not approach the meeting point without some way to protect yourself. If you are not comfortable going to the scene, stay away until you are given an assignment you are comfortable performing.

- In some cases, it may be possible for animal staff to gain animal containment using positive reinforcement training techniques. These plans should be discussed with the on-duty IC, Animal Care Manager and/or Recapture Coordinator.
- The on-duty IC, firearms team, chemical immobilization team, and security officer should be the only units to acknowledge the call, with all other units assuming radio silence.

Animal Escape Occurring While Non-Animal Care Staff is on Property Involving Dangerous Animals

If the Code occurs while people other than animal care staff are on property and involves a dangerous animal, the IC should direct employees to seek shelter for guests and themselves in safe areas such as buildings, or night houses that are safe or have an animal care technician area. In some cases, it may be necessary and safer to have

guests and non-animal care staff exit the facility if doing so does not provide a way for the animal to leave zoo property. A staff member should be present with guests until the code is resolved.

Incident Commander Responsibilities

- Work to provide for the safety of everyone on property by making radio calls to move the non-essential staff and the public to indoor safety. They should direct the firearms team and liaison with outside agencies if needed. They should work with the Recapture Manager to ensure the safety of all people and animals.
- Give instructions to close all perimeter gates and service gates to confine the escaped animal within the zoo grounds. Certain gates should have staff in charge of securing the specific gates. The staff assigned should be close to the gate as a normal part of their routine and not involved in the firearms response team.
- Ensure firearm equipment will be transferred to the site if needed. Only authorized and trained firearms team personnel will use firearms equipment.
- Ensure chemical immobilization equipment will be transferred to the site if needed. Only authorized and trained immobilization personnel will use chemical immobilization equipment.
- Assess the number of people needed to assist in capturing the escaped animal and designate a meeting spot.
- Notify the veterinarian on duty or call the contract veterinarian to come to the facility. In some cases, the fewer people involved the better. The IC should work with the Recapture Manager to ensure that appropriately trained staff are responding and staff responding is trained.
- Notify the Head of Animal Husbandry and Chief Animal Officer.
- The Head of Animal Husbandry will notify the IACUC and any other pertinent Colossal personnel.
- Ensure that a debriefing meeting is conducted within 24 hours, **including a Colossal husbandry team member**, and should go over what went well, what can be improved in the future, and any other lessons learned.

Emergency Response

Emergency Contact Name and Title

Cell Phone Number

Curator/Animal Director:

Security:

Firearms Team:

Local Police Department:

Additional Local Police Department: Local Fire Department: Additional Local Fire Department: Local Ambulance Service: Local Poison Control Center: Colossal Head of Animal Husbandry: Colossal Manager of Animal Husbandry:

Emergency Response Actions

Emergency response actions should be taken immediately upon determination of an impending disaster or emergency related event in effort to prevent loss of life and alleviate suffering. The more familiar staff are with what to do in an emergency, their roles in the emergency, and how to prepare and respond to will help determine the outcome. The more prepared the staff the better the outcome. Staff should be familiar with the following procedures and adapt these procedures to emergency situations that arise. No document can cover or prepare staff for every possible situation, but this provides a foundation to handle sudden, impending, and unexpected situations that could potentially become emergencies.

Drills are conducted at a minimum of once a year. Drills may include but are not limited to tabletop discussions and in the field practice scenarios. All drills will be documented and stored in accordance with all accrediting organizations requirements. Drills will be conducted for all scenarios including equipment at least once a year to ensure a swift and accurate response to an emergency.

If an outside credible threat is identified, the threat will be evaluated by the Head of Animal Husbandry and an appropriate increase in security will be deployed and appropriate entities will be notified.

Temperature requirements listed in this SOP do not replace temperature guidelines defined in the <u>Environmental</u> <u>Parameters</u> and <u>Neonate Rearing Guidelines</u>.

Barrier Breach

Animal escapes are addressed in the <u>Animal Escape SOP</u>, but if a person should breach the animal's primary containment and be inside with Colossal dire wolves and their life is in imminent danger, certain precautions and decisions will need to be made quickly and action taken. In this instance, the senior staff member or security will make the decision and let the Head of Animal Husbandry know the outcome to ensure human survival. The order of concern is to ensure community safety, ensure staff safety, and then animal safety. If a human has breached the animal's habitat and their life is in danger:

• Remain calm.

- Security, firearms team, or law enforcement should be immediately contacted and told to be ready to destroy the animal at the instruction of the senior most animal personnel or Incident Commander.
- Staff should immediately attempt to use extreme measures to separate the human and the animal (e.g., loud noises, hoses, foreign objects, etc.). Staff should not go into the enclosure or put themselves in danger.
- Staff should attempt to recall the animals off exhibit.
- When on-site security, firearms team, or law enforcement arrives, if the animal has not been successfully separated from the human, the senior most animal personnel or emergency coordinator should give the instruction to destroy the animal. The senior most animal personnel or veterinarian will be responsible for reporting the incident to the Head of Animal Husbandry. The Head of Animal Husbandry will be responsible for reporting the incident to the IACUC and other appropriate Colossal staff.

If a human breaches an exhibit barrier and their life is not in imminent danger, certain precautions must be taken:

- Security, firearms team, or law enforcement should be immediately contacted and told to be ready to destroy the animal at the instruction of the senior most animal personnel or Incident Commander.
- Staff should make the attempt to extract the human from the enclosure without putting themselves in harm's way. Staff should never enter the exhibit with the animals.
- Staff should attempt to recall the animals off exhibit.
- Staff should immediately attempt to use extreme measures to separate the human and the animal (e.g., loud noises, hoses, foreign objects, etc.).
- When on-site security, firearms team, or law enforcement arrives, if the animal has not been successfully separated from the human, the senior most animal personnel or emergency coordinator should give the instruction to destroy the animal.

After any incident, there will be an immediate staff briefing on the procedures and anything else pertinent to this subject. There will be an additional meeting on how to prevent the situation from occurring again.

Fire on Grounds

Prevention is the best protection against fire. Faulty wiring, exposed wires, and problems with heaters should be reported immediately and not used until inspected and fixed. Heaters, especially supplemental heat lamps and heat blowers should be checked regularly, secured properly, and staff should check them daily to ensure no flammable materials are near them. Flammable materials and items should be stored properly and kept clean. Heaters and filters should be cleaned regularly and have a maintenance schedule that is adhered to. Any concerns should be discussed with management and an appropriate maintenance person should be contacted to address the situation.

Each building is equipped with Class B and Class C Fire Extinguishers (Class B extinguishers for fuel fires such as gasoline, oil, and propane; and Class C for electrical fires). Some extinguishers are class ABC that work for wood and paper as well. BCs do not work for burning solid materials such as wood and paper. Class A's or water cans do not work for fuel or electrical fires. All extinguishers should be checked annually. When using a fire extinguisher, remember to use the appropriate class for the type of fire and remember the PASS method:

Pull the pin Aim to the base of the fire Squeeze the handle

Sweep the area

If a fire is observed on property or near property, staff members should report the fire to their supervisor and include the following information:

- Specific location on property
- Type of fire: building, dumpster, grass, electrical?
- What animals and people are in the structure or immediate area?
- If it's a building or barn, have people been evacuated or are they evacuating?
- Is anyone or anything in need of medical assistance?
- Has 911 been called?

If a fire extinguisher is close by and it is safe to do so, staff can use the fire extinguisher and put out the fire. Staff should be trained to use the fire extinguishers during their new hire training. They should know the difference between an ABC and Co2 extinguisher and use the appropriate one for the fire. If animals can be safely evacuated, do so and shut them out of the building. If a fire is too large and/or can't be extinguished with a fire extinguisher, evacuate the area, closing the doors behind you. Immediately notify the senior animal care staff. The most senior animal care supervisor will head to the site of the fire, announce their presence via radio and text message, and be the Incident Commander.

Incident Commanders will assess the situation as quickly as possible and coordinate a response effort, communicate with Colossal, and delegate as needed. The responses may include some or all of these and are not limited to the following:

- Ensure 911 has been called and given the appropriate and specific information of the emergency, and the closest entrance into the facility to the fire.
- Evacuate all structures, closing doors behind you as you leave, evacuate the surrounding areas of humans, consider which way the wind is moving or pushing the fire, and notify the areas of the safety risk.
- Set up and maintain a perimeter.
- Communicate medical attention needed and get the appropriately trained staff to respond until the first responders arrive.
- Have an employee station at the entrance with the gate open unless there is an animal escape in which case the employee should open the gate when the fire engines approach if the animal is not in the area. Another employee should be with them to lead the engine into the facility.
- Determine if there are animals at risk in the area or building and if so, work with a senior member of the animal care department referred to as the Animal Commander (AC from this point forward) familiar with the building and animals to assess the escape risk and if evacuation is possible. The Incident Commander and the AC should stay near each other to communicate directly to ensure the best and safest outcome.
- Animal care technicians will report to the Animal Commander and be instructed by the AC to receive information and instructions as to evacuation and fire response.
- The IC and AC will develop an animal evacuation plan and adapt it as the situation changes. Staff, equipment and tools, and any veterinarians will be just outside the perimeter to receive evacuating animals and work with the first responders when they arrive.
- Animal evacuations should not occur if it is unsafe for staff to do so. The Fire Department Incident Commander will be made aware of all animal-related issues.

• Animals may be shifted within their enclosure/barn/night house areas unless they are in a species specific secure and approved transport crate or trailer. Before moving the animal, a different enclosure and holding facility must be available as the animal cannot live in its crate long term, and its original primary location may be destroyed. No animal should be put in a crate, trailered, or moved without the authorization of the Head of Animal Husbandry, senior management, and the IC. This should be determined before any emergency and be part of the plan. Quarantine facilities may be used, including other facilities in which the original facility may offer temporary suitable housing. Animals may be shifted to outdoor yards and enclosures if safe and don't pose an escape potential.

Fire After Hours

If staff live on-site, they will be notified immediately by those monitoring the surveillance cameras. Once notified, staff will make their way over to the scene of the fire and assess the area containing the fire. If unable to contain the fire, the appropriate immediate action must follow. If you can safely extinguish the fire, locate the fire extinguisher and do so. If you can safely evacuate the animals from the building, do so, closing the doors behind the animal. If you cannot control the fire, close the doors to try and contain the fire.

The camera monitoring desk can be contacted via phone or radio. If you are the first one to notice the fire, contact them and they will guide the fire department on-site and directly to the fire.

Phone # of staff monitoring cameras:

Radio # of staff monitoring cameras:

There will be an immediate staff briefing on the fire, the procedures, and anything else pertinent to this subject. This will be led by the senior most member of the animal care team.

Wildfires

Wildfires will be handled based on the direction and speed of fire, and communication from the Fire Department or the wildfire Incident Commander. The location, size of fire, road conditions, etc. will influence the course of action. If animals can be moved into their crates, or trailered to safety and there is time to do so, the Incident Commander and the Head of Animal Husbandry will determine if that is the best course of action. Priority will be given to staff safety and ensuring animal containment. This means that some animals may be secured in their buildings and not moved.

- Bring animals into their barns or crate the animals and load them onto trucks/trailers to be moved to a safer location. All large carnivores must be secured in a building or crated and moved if there is time. Large carnivores cannot be allowed to escape containment and pose additional emergency threats to the facility and community.
- Some animals may be let out into large acre pastures to allow them to try and stay out of the path of the fire if they are in a large acre facility. This will depend on if they are dangerous animals, the area, and recapture possibilities. Fences between pastures may be cut to allow the animal more area to maneuver around the wildfire. Perimeter fences should never be cut. Plywood can be placed across cattle guards to help animals move to safety.
- If time allows, shut doors and windows to buildings and shut off natural gas. Turn off electricity and heating and air conditioning to prevent electrical fires inside structures.

- Hose down structures and surrounding areas if there is time. Sprinklers should be turned on.
- Move vehicles and equipment out of the path of the fire or to a location further away from the direction of the fire.
- Move portable propane tanks, fuel cans, and oxygen and welding tanks out of the path.
- Wet plywood should be placed over vents and windows if time.
- Remove brush, trees, and other flammable debris from around structures and perimeter of the zoo. This should be a part of regular maintenance, but distance may be increased if wildfire threat is present.
- Provide animals with large supplies of freshwater. Fill extra bowls, troughs, buckets, and containers with fresh drinkable water.
- Remove all firearms and ammunition.
- Veterinarians or members of the chemical immobilization team should have the dart gun and chemical immobilization drugs available to them in case of an animal escape.

The Head of Animal Husbandry should provide a list of support animal facilities that should be notified and can help in case of wildfire and extreme weather. The list should include what species they can house. Multiple facilities should be identified and listed. This should include transportation assistance such as trailers, tractors, stock trailers, and food suppliers. You will need to have resources to move, house, and feed the animals for the long term if you have enough time to move the collection. You will also want to be able to feed your staff if they are evacuating animals. These facilities should have an MOU in advance and a mutual agreement of assistance be solidified in advance of the emergency.

If the facility is in a voluntary evacuation zone, preparations should begin immediately if they haven't already started. Animals can be crated and staged in a centralized location. Ungulates can be trailered and moved to other locations if they don't have crates. Staff members required should be present, and if needed, alternate staff that may be able to step in if a required staff member needs to evacuate themselves and or their dependents. A ride out crew should be designated in advance to stay on the ground in case the situation continues to develop, and animals need to be evacuated. Staff should be rotated out to prevent fatigue. A senior member should always be part of the ride out crew. The Head of Animal Husbandry, a supervisor, and a veterinarian will determine which animal will be evacuated as soon as the wildfire is in the known area. Staffing will also be determined based on the number of animals, the species, and the skills of the team. Staff will need to be positioned and available at the holding areas to care for the animals during transport and staging. Large taxa may have to shelter in place. This will be decided in advance, and monitoring of these animals will happen from remote cameras while their ability to do so is available. The ride out crew will need to let the fire department know their location and may have to evacuate if fire reaches the property.

The Head of Animal Husbandry, senior management, and the veterinarian may decide that animal euthanasia may need to be deployed to prevent an unavoidable and painful death due to fire related health problems. This will be decided by the team and conducted by the veterinary staff or other appropriately trained staff member.

Inclement Weather/Natural Disaster

Animal and personnel safety is number one priority. To ensure both the safety of all animals and the safety of all personnel and its neighbors, weather will be monitored constantly.

All Colossal dire wolves will be locked into their nightly quarters if any major storms are to pass through the area such as, but not limited to thunderstorms, snowstorms, hurricanes, tornadoes, heavy rain, flooding, heavy wind, etc. If there is any question as to what is considered as inclement weather, please contact the animal care manager.

Colossal dire wolves can have access to their outside yards in almost any temperature, but precipitation and wind should be monitored constantly to determine if animals can go out for the day or should be brought in early due to impending inclement weather.

If there are winds over 30 MPH, heavy precipitation, and any weather with poor visibility, all animals should be locked inside until exhibits can be checked and deemed safe and ready for animals to go outside.

After any storms, exhibits will be thoroughly checked for any downed trees, breaches in primary and secondary containment, functioning hotwire (5V or higher), unsafe conditions, etc., before shifting animals outside. If anything needs repair, a manager should be notified immediately, and animals will be kept inside until the necessary repairs are completed. Once the exhibit is deemed safe, animals may resume normal daily operations. Trees are visually inspected daily, however a more thorough inspection occurs annually by a horticulturist to check tree health.

All buildings are connected to a generator that will automatically switch power over whenever power happens to go out. If for whatever reason the generator does not kick on, flashlights and headlamps are housed in the buildings and garage doors can be manually opened to let natural light in if needed. Freezers and refrigerators will not be opened to maintain temperatures. If power is out for more than six hours, frozen and refrigerated goods should be moved to an area on the farm that has a backup generator. If the generator does not kick on, notify management immediately and do not let animals out until hot wire is back on (recall animals if needed).

Inclement Weather Overnight

All dangerous animals will be locked in their respective holding areas. Many staff live on-site and will do regular fence checks throughout any inclement weather events if it is safe to do so.

Flooding

When heavy rainfall or flooding is predicted for the area surrounding the facilities, staff should clear any drainage ditches, drain systems, culverts, or water gaps. During and after rain events, staff should inspect the drainage sources and remove debris if it is safe to do so. Water moves fast and at no point should staff place themselves in danger to clear debris from a waterway. Water often can move faster below the surface. Pool skimmers can be used to remove small debris, and when it's safe, heavy equipment may be used to remove big debris. Staff should continuously monitor fence lines of the perimeter and of the enclosures for fallen limbs and fence damage. Animals should be secured inside their indoor holding areas and if needed, crated/trailered to higher ground if the building is in threat of flooding. The Head of Animal Husbandry should be made aware of the situation when a heavy rain event is predicted. If a building is in a potential flood area, move any items up off the floor by at least two feet and make sure any electrical outlets are shut off.

High Winds and Thunderstorm Warnings

High winds (winds over 30 mph) create hazards for falling tree limbs, falling trees, and flying debris. Care should be taken to maintain and prevent tree limbs from having the potential of falling on power lines, power boxes, and fence lines. Regular walking/driving of enclosure fence lines and perimeter fence lines should be done as part of the animal care technicians' daily duties. Each area animal care technician is responsible for

their enclosure fence lines and monitoring of trees, informing facilities management and maintenance teams, and recording potential and existing issues on the animal's daily record in the animal care information management system. The Head of Animal Husbandry will be notified by the area supervisor of potential hazards at the weekly check-in meetings and by reading animal care technicians' daily logs. After a wind event, fence lines should be walked/driven and monitored for integrity if it is safe to do so. Staff should wear eye protection to prevent injury from flying debris. If tree limbs and trees need to be removed, only staff specifically trained to use the equipment needed may use the equipment. Staff should wear the appropriate PPE when operating equipment and completing potentially hazardous duties.

All Colossal dire wolves should be locked inside during high winds to prevent escape potentials that may be caused by falling debris. They should remain locked in until the integrity of the fence lines and nearby trees are assessed.

After high wind events, all pools, troughs, and drains should be skimmed or cleared of debris. Waterways and walk paths should also be cleared of any fallen debris.

Extreme Storm Events: Hurricanes/Tropical Storms/Tropical Depressions

When the path of a hurricane is predicted to impact the facility, staff should advise the Head of Animal Husbandry as soon as possible. The supervisor and Head of Animal Husbandry will create a plan for the specific facility and species. They will set priority for animals to be brought in and determine where they will be housed. They will determine and delegate the ride out teams. As items are completed, the Incident Commander should be notified and is responsible for maintaining and recording completed tasks. Due to the amount of time to prepare for hurricanes, there may be a pre-storm IC and an IC for the ride out crew.

The ride out IC will determine when and if it is safe for staff to check on animals. The eye of the storm usually allows time for checks of animals and grounds to be performed. Once the storm and storm threats have passed, the ride out IC and ride out crew will stay until the non-ride out crew can arrive. Animals' habitats will need to be fixed and ensured secure before animals can be released from their hurricane holding areas. The IC will be responsible for notifying Head of Animal Husbandry of animal, staff, and facility updates and incidents.

Storm is 48+ hours away:

- Area should be cleared of debris that may turn into flying objects.
- Prepare barns/shelters for long-term holding of animals.
- Make sure all emergency equipment and generators are fueled up and working.
- Additional supplies such as food, drinkable water, and fuel should be procured.
- Start moving items off the floor, they should be at least two feet above the ground.
- Start covering windows with plywood.

Storm is 24-36 hours away:

- Secure equipment.
- Stock barns with containers to hold drinkable water. These can be stored in extra troughs, bowls, jugs etc.
- Stock emergency supplies.
- Fuel up all vehicles and move them to secure locations.
- Clean all bedding out of stalls and drain baskets out of drains, leave drain covers on, and mark storm drains with stakes as the drain might not be able to be seen in high water.
- Put together and collect any capture equipment needed.
- Ride out team members should be notified. A senior member of the animal care team and a veterinarian or trained member of the chemical immobilization team should be on the ride out team.
- Make sure flashlights, back up chargers, and generators are charged, fueled, and working.
- Start cleaning all areas and preparing animals, stock feed and fill up any food containers, stock supplies for several days.
- Get food and water ready for the ride out crew.

Storm is 12-24 hours away:

- Secure records in a waterproof safe area.
- Drain the animals' outside pools.
- Finish boarding up windows and reinforcing caging.
- Get out heavy rain gear, waders, and other PPE for the ride out crew. The ride out crew should be home preparing at this time and should arrive 12 hours before the storm is estimated to arrive.
- Bring animals into their respected barns and holding areas that are designed and determined to be storm safe.
- Disconnect electric fences. Manually lock all internal and external gates with chains and locks.
- Shut off the hotwire.
- Disconnect all non-essential electronics. Cover computers and other electronics.
- Sandbag doors to barns and food storage areas.
- Fill coolers and troughs with ice or drinkable water.

Storm is 6-12 hours away:

- Ride out crew arrives, and non-essential personnel are released to go home.
- Secure vehicles in the building and leave one vehicle outside the door of the staff ride out shelter. It should be a 4x4 truck with keys in it and doors unlocked.

Storm is imminent:

- Shut down and secure all areas.
- Staff reports to ride out the shelter.
- Report to the IC as areas are shut down and secure.
- Report to the IC when each staff member arrives at the shelter.

Tornadoes

If there is enough warning, all Colossal dire wolves should be secured inside their buildings. This should be done where the thunderstorm or high wind (30 mph or higher) event occurs, which usually but not always precedes tornado watches and warnings. However, some tornado events occur with very little warnings and if that occurs, staff should find a safe place to shelter and deal with the events after. Staff should prioritize their safety over securing animals if a tornado is in the vicinity. If storm shelters are nearby, staff should get to them. If staff are not near a storm shelter, a ditch or culvert that isn't a flood risk should be found. Staff should not

seek shelter in vehicles and should know which buildings are qualified to be storm shelters, especially where tornadoes occur. Once the storm has passed, staff should check and verify locations of animals, fence line integrity and security, and notify the senior member of management. The supervisor should be in contact with the Head of Animal Husbandry as soon as possible to advise them of the situation. Staff should try to secure animals into their holding if their holding is safe and structurally sound to hold the animals. The Incident Commander will work to assess the facility and animals after a tornado.

Animal Evacuations

In most natural disaster cases, it will not be possible to evacuate all the animals due to time, safety, and logistical issues. A list of animal-related facilities in the area should be kept and maintained that may be able to temporarily help house some of the animals in the facilities' collection while repairs and recovery efforts are made. Facilities should look into Zoological Disaster Response, Rescue, and Recovery (ZDR3) before disasters strike as they can provide assistance before and after a disaster. The Head of Animal Husbandry and senior animal management will work together to make decisions regarding Colossal animals. The Head of Animal Husbandry and the veterinary staff will be responsible for making decisions on anesthetizing and/or euthanizing animals which have been injured due to an emergency or natural disaster or preventing serious injury that could result from an impending natural disaster or emergency.

Power Outages

The facility has backup generators for any containment doors that are electric. Backup generators should be employed if there are power outages during extreme cold, such as extreme arctic storms where animal life or quality of life can be compromised by extended exposure to sub-freezing temperatures. Even though the grey wolf has evolved to endure the harsh winters of northern climates, they are not able to freely move and choose better spots to help stay warm in all the ways they have evolved and are not kept in large packs to help with warmth. Water should be put into containers and stored to ensure that if the power goes out and well water or pumps are affected, animals will always have access to water.

Arctic Blizzard or Extreme Sub-Zero Temperatures

All buildings should have whole house backup generators to maintain lighting and electrical capabilities. This will help maintain temperatures in their buildings and will help to keep pipes from freezing if it becomes cold enough where the facilities' pipes are in danger of freezing. Facilities should follow any emergency procedures recommended by local authorities. Some may include leaving a trickle of water running from faucets or hose bibs. Colossal dire wolves may be given access to the outdoors but must not be locked out in temperatures that are below 32°F for a length of time other than it is necessary to provide essential husbandry. If temperatures are near or below 0°F, care should be taken to not lock any Colossal dire wolf outside for any length of time. Colossal dire wolves should be provided additional bedding if high temperatures are not going above zero. Depending on the temperatures, the Head of Animal Husbandry and senior animal care supervisors will decide if changes are needed to daily husbandry such as additional heat, no hosing of floors, etc.

During blizzards, staff should monitor and check fences for heavy ice or snow that may compromise the integrity of the primary and secondary containment and attempt to remove the buildup of snow/ice. Staff should check yards and areas above shift doors, trees, and fences for icicles that can fall and impale Colossal dire wolves or staff and knock them down before they become a hazard. Heavy snow and ice can pull down trees and tree limbs just as easily as fences. Staff should monitor and rectify any safety concerns before Colossal dire wolves are given access to the area or recall the Colossal dire wolf as soon as possible to prevent injury or

escape. Animal care staff may need to dig out shift doors and animal care technician doors to safely open and close doors during a blizzard. Snow can collapse and prevent shift doors from closing or doors can become frozen as snow and ice melt and refreeze due to the warmth of the inside barn areas. Staff may need to put on tire chains to ensure they can get to buildings to care for animals.

New Staff Training

Animal care staff will be responsible for training existing and new animal care technicians in all aspects of being an animal care staff for Colossal dire wolves on property. This SOP has been adapted and modified by Colossal for use in training staff with the Colossal dire wolves.

Only trained animal care staff will be working with and providing daily husbandry for Colossal dire wolves. These operating procedures are to be trained to and used by all animal care staff providing daily husbandry. Any exceptions must be approved by the Head of Animal Husbandry. Staff other than an animal care technicians trained to care for the Colossal dire wolf, will need to be discussed with the Head of Animal Husbandry in advance.

The daily husbandry of the Colossal dire wolf collection must exceed the standards outlined on the AZA website to "ensure that the physiological, biological, psychological, and social needs of the animals' are always met. Using nutritionist prescribed diets, enrichment, cleaning, health checks, and training of animals/staff, the husbandry requirements of Colossal Biosciences and the AZA should be met daily.

Animal care staff and supervisors are responsible for ensuring that the highest standards of care are met. The staff members must keep daily records in the animal care information management system of animal health/ behavior, diet, training, and enrichment.

During training, new staff will be trained how to safely care for the animals including but not limited to: how to observe and monitor animal behavior for any health issues, how to safely work around and shift the animals, diet preparation, how to input records, how to properly medicate animals, how to safely provide enrichment and training, and anything else regarding the care of Colossal animals.

Safety

• All staff will be appropriately trained and prepared to respond to emergencies (fire, inclement weather, floods, power outages, animal escape, natural disaster, etc.). See the following SOPs for detailed instructions and guidelines:

- o <u>Emergency Response</u>
- o <u>Environmental Parameters</u>
- o <u>Animal Escape</u>
- o <u>Shifting Procedures and Precautions</u>
- Individuals involved in the care of animals must use safety tools, procedures, and good judgment to maintain the health and safety of all animals and persons.
- See species specific Colossal Dire Wolf SOP/husbandry manuals for safe handling instructions.
- Staff will all be trained and aware of safety procedures to minimize/eliminate risk of zoonotic disease.
- Any exposure to animal bites, scratches, stings, etc. must be reported in an incident report submitted to appropriate management within 24 hours.

Training Colossal Dogs (Non-Dangerous Animals)

This may include dire wolf pups with surrogate dog dams. All animal care staff being trained on a new shift will undergo a 2-week training process, with the exception of dangerous animal training (4-week training process).

- Day 1: A shadow day is instituted so new staff can obtain a brief but thorough overview of the new routine. Ideally this day should be led by a senior animal care staff. New staff should be trained on proper radio etiquette and be given access to read all Colossal Policies and SOPs.
- Days 2-3: After a full shadow day occurs, the new staff member will lightly begin assisting the senior animal care staff with shift duties. Day 2-3 is still solely led by senior animal care staff. Advice and insight can and should be given throughout this day.
- Days 4-6: The new staff member should start to be able to take on some tasks on their own during the day. Senior animal care staff should be alongside the trainee every step of the way and continue to be direct with instructions.
- Days 7-8: Starting and finishing all daily tasks should start to become routine for the trainee. While senior animal care staff should be by their side all day, the bulk of the work is to be completed by the new staff member. Senior animal care staff should also be asking for the trainees' comfort level with the animals and time management at this point.
- Days 9-10: The final days should be started and finished solely by the new staff member. At senior animal care staff's discretion, they can be used as a "satellite animal care technician" or can shadow the trainee but allow the new staff member to complete the final day on one's own.

Training Milestones

- The trainee should take note that training sessions and major animal shifting should only be done by senior animal care staff in that area until approved by a supervisor.
- If there are any concerns about "shift clearance," a manager should be contacted immediately.
- There may be information that will still need to be learned that a new staff member may not have seen during their training in a section (e.g., rotational feed schedules or cleaning schedules that happen on a day the new animal care technician did not train in the area).

Training Colossal Dire Wolves–Non-Domestic (Dangerous Animals) Over 45 pounds

• All staff members being trained on a new shift will undergo a 4-week training process when working with dangerous animals including dire wolves. During this 4-week period the trainee should learn the natural history of the closest relative(s) of the Colossal dire wolf they are working with. They should

learn the history and behaviors of the species, and closest members of the species. This will help new staff to interpret interactions and behaviors accurately and to ensure accurate and important information is recorded and passed on so that management decisions are made to optimize animal welfare.

- Trainees need a minimum of four weeks working with senior animal care staff of Colossal dire wolf areas.
- Trainees must shift with all members of the Dangerous Animals area team, or all trained members of the area Colossal dire wolves are housed.
- The senior animal care staff and the trainee must agree that the trainee is ready to move on to each new step of the training process (this timeline can be delayed if needed).
- All aspects of the routine should be checked off before the trainee can do tasks on their own (Colossal suggests following a training check off list to ensure trainee is trained on all necessary duties).
- New trainees will start operating shift doors no earlier than the 2-week mark with senior animal care staff present.
- If any staff member feels uncomfortable with a trainee at any point in the training process, management is notified, and training stops until further notice.
- A meeting will then be held with the trainee to explain why training stopped and if training will continue in the future.
- Trainees are expected to communicate appropriately such as, but not limited to, speaking clearly, loudly, and using the appropriate dialogue and abiding by the only activities required for shifting animals. No chit chat, phones, watches or any other non-shifting communication or activity is permitted.
- Staff should be 100% present in the job they are doing to ensure the animals get the best care and all people and animals are safe.
- Trainees are required to fully understand how to manually use shifts and the layout of the Colossal dire wolf buildings.
- Trainees will be considered fully trained once all DA staff and management have signed off that the trainee is cleared as a shifter and has been observed shifting in all areas by management.

4-week training Process

- Day 1: A shadow day is instituted so the new staff member can obtain a brief but thorough overview of the new routine. Ideally this day should be led by senior animal care staff. New animal care staff should be trained on proper radio etiquette, pepper spray usage and be given access to SOPs for the area.
- Week 1: After a full shadow day, and all Colossal Policies and SOPs are read and checked off, the new animal care technician will lightly begin assisting senior animal care staff with shift duties. Senior animal care staff should also be asking for the trainee's comfort level with animals and time management.
- Week 2: The new animal care technician will physically learn how to operate shift doors in a controlled animal-free environment with senior animal care staff before advancing to actively shifting Colossal dire wolves. Senior animal care staff should be alongside the trainee every step of the way and continue to be direct with instructions and asking the trainee's comfort level with the animals and time management.
- Week 3: Trainee is shifting dire wolf while shadowed by each member of the respective trained member of the dire wolves' care team. Finishing all daily tasks should start to become routine for the trainee. While senior animal care staff should be by his/her side all day, the bulk of the work is to be completed by the new staff member. Senior animal care staff should also be asking for the trainee's comfort level with animals, shifting, and time management.

• Week 4: The final week should be started and finished solely by the new animal care staff. At the senior animal care staff's discretion, they can shadow the new animal care staff but allow the trainee to complete the final days on one's own. Training is concluded with management shadowing the trainee shifting in all dire wolf areas and signed off by all trained members of the dire wolves' care team, and Head of Animal Husbandry or Manager of Animal Husbandry.

Colossal Dire Wolf Training Milestones

- The trainee should take note that training sessions and major animal shifting should only be done by a primary animal care technician in that area until approved by a supervisor.
- If there are any concerns about "shift clearance," a supervisor should be contacted immediately.
- There may be information that will still need to be learned that a new staff member may not have seen during their training in a section (e.g., rotational feed schedules or cleaning schedules that happen on a day the new animal care technician did not train in the area).
- **This SOP exists in a vacuum and does not account for such things as vet visits, mandatory meetings, emergencies, and learning styles, all of which occur regularly, disrupt training, and vary based on situation and person. Notify the supervisor immediately if these tasks conflict with your ability to learn and or accomplish your daily job.

Literature Review

REVIEW OF LITERATURE THAT ALSO UTILIZE CANIS LUPUS FAMILIARIS (DOG) AS SURROGATES

There has been extensive research in breeding and assisted reproduction in Canis lupus familiaris that demonstrate why they are good candidates for surrogacy.

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Website Resources:

Association for Behavioral Analysis International

www.animalenrichment.org

Cambridge Center for Behavioral Studies

B F Skinner Foundation

AZA animal training guiding principles

AZA Enrichment guiding principles

AZA/AAZK Animal Training Terms and Definitions

AZA Animal Training Recommended Resources

AZA training Session Feedback Form Accompanying Guide

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Nutrition Advisory Group Association of Zoos and Aquariums Santa Barbara Zoo Disney's Animal Kingdom Phoenix Zoo San Diego Zoo Wildlife Alliance Oakland San Francisco Riverbanks Zoo EL Paso Zoo Oklahoma Zoo Dallas Zoo