

Positive-reinforcement strategies to reduce capture-stress in Capybaras

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ABSTRACT. Prior to the administration of any anesthetics, capturing the semi-aquatic capybara (*Hydrochoerus hydrochaeris*) demands physical restraint, which presents immense challenges. Traditional methods, such as lassoing and traps, are prone to induce acute stress, may result in injury, and can even have fatal outcomes. As part of a larger population control project using contraceptive methods, frequent capture-induced stress and injury may directly affect normal reproductive physiology. Thus, choosing a less stress-inducing method was imperative. In this report, we describe methods of conditioning to enable frequent capture and manipulation, using bait as a positive reinforcement associated with a special click-sound, in a free-ranging population of 40 capybaras. The objectives were to attract, herd, and allow capybaras to voluntarily enter a coral. We evaluated the conditioning effect on individual and group behaviors, interpreting vocal and body language manifestations during the processes of conditioning, herding, capture, and recovery (post procedure/chemical restraint), with the aim of minimizing capture-related stress and injuries. Based on our observations, we report that conditioning, used as part of the capture strategies, noticeably facilitated physical restraint and manipulation throughout the procedures, while apparently maintaining the animals' overall welfare as it relates to conditioning and capture.

Keywords: animal welfare; baiting; conditioning; *Hydrochoerus hydrochaeris*; physical restraint; behavior.

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Introduction

Capturing wildlife species for research is a frequent necessity and is often accompanied by great challenges; as is the case with capybara (*Hydrochoerus hydrochaeris*). The capybara is the world's largest rodent and native to South America. It has an average adult weight of 40kg, and in its natural habitat may live in groups of 5-20 animals (Alho & Rondon, 1987). This is different to synanthropic capybaras (Figure 2), which re-inhabit either agricultural or urban areas and have body weights that can reach over 100 kg. In addition, because of their high proliferative nature, groups can quickly reach large numbers, with over 100 members in some cases (Labruna, 2013; personal communication; personal observation, 2018). This is largely due to a lack of natural predators, the abundance of food, resistance to anthropogenic activities, and their ability to quickly adapt to urban dynamics (Marchini & Crawshaw, 2015).

One of the main reasons why studies on capybaras have increased is because of their potential threat to human health, being an amplifying host of *Rickettsia rickettsii*, the etiological agent of Brazilian Spotted Fever, a lethal tick-borne disease (Fiol, Junqueira, Rocha, Toledo, & Barberato Filho, 2010; Labruna, 2013). Thus, finding the most adequate method of population control, concurrent with collecting material for epidemiological surveillance, has become a top priority (Souza, 2004; Labruna, 2013; Felix et al., 2014; Bovo, Ferraz, Verdade, & Moreira, 2016). Controlling capybara populations is limited to non-lethal methods, pertaining to the native fauna, they are protected by law from hunting, trafficking, and abuse (Brasil, 1967).

Capybaras live in social groups composed of one dominant male (alpha), subordinate males, adult females, juveniles, and infants. Sexually mature males, expelled from the group by the dominant male, so-called satellite males, are commonly observed in close proximity to the group (Figure 1). The presence of these satellite males may lead to group division when females abandon their group (Macdonald, 1981), or they may simply assume leadership if the alpha male is injured, ill, or dies.

Capybaras are a semi-aquatic prey species for large felines, alligators, and anacondas. Besides using water for hydration, thermo-regulation, copulation, and defecation, it serves as the principal getaway in

case of a predator attack (Moreira, Ferraz, Herrera, & Macdonald, 2013). Capybaras are quite fast and agile animals; however, their stamina is short-lasting, which is why they stay in close proximity to water (Figure 2). As such, capturing capybaras, without putting their lives at risk, is notoriously difficult.



Figure 1. *Raia Olimpica*, Campus University City, *Universidade de São Paulo*, Brazil; Green arrow, pointing to a satellite male, close to the water, identified by secondary sexual characteristics of an adult male, solitary position, and through prolonged observations; The



white arrow points to a group of about 30 members, crossing the water.

Figure 2. Google map adapted; a) South America; b) Metropolitan area of São Paulo; c) Bird view: *Raia Olimpica*, a man-made water pool for aquatic sports; d) Group of synanthropic capybaras, close to the water.

The literature describing detailed capture methods for capybaras is limited and the few studies that are available are not suitable for an English-speaking audience. However, capture methods can be divided into three groups: (1) lassoing, a traditional method commonly employed by Argentinian cowboys (Salas, Pannier, Galíndez-Silva, Gols-Ripoll, & Herrera, 2004); (2) baited heavy metal traps; and (3) corrals with

bait, as described in several research projects (Souza et al., 2008; Moreira et al., 2013; Kuniy et al., 2018). The first two methods can potentially provoke intense stress and have an increased risk of injury or death and would make recapture almost impossible. Logistics are a great challenge when frequent capture is necessary, particularly for semi-aquatic capybaras that require prior physical confinement, as any anesthetized animal would almost certainly flee into the water, if not physically restrained, and inevitably drown after the onset of the anesthetic drug (personal communications, 2017). Furthermore, it is important to use capture methods that allow the capture outcome to be more controllable (targeting individuals or larger number of members, time independent), rather than relying on random capture using baited traps.

As part of a greater research project on population control using immunocontraception in free-ranging capybaras (São Paulo, State, Southeastern Brazil), we investigated different capture methods that would allow for frequent selective capture, throughout the year. Capybaras have a keen olfactory system when it comes to detecting food, especially sugary plants. Observing their attraction to specific foods, curiosity, and gregarious social structure, we recognized the potential for attracting, and even training, capybaras using methods based on classical and operant conditioning, reasoning that it would aid our overall project and allow for frequent physical restraint but minimize capture-related stress and injuries.

Briefly, the concept of classical conditioning (or Pavlovian conditioning), takes advantage of an unconditioned stimuli (food) that is natural to the target species, in case of capybaras, sugarcane. Once attracted by the smell and voluntarily approaching the food source, referred to as the unconditional response (not based on behavior), food offerings are accompanied with an easy to recognize sound. This subsequently creates a classical conditioning scenario by combining the unconditional response with a conditioned stimulus, which over a short period of time, turns into a conditioned response, providing the fundamental principles to expand to operant conditioning (or, Skinner Principle). For example, motivated by the click-sound and lured by the bait, the group could be round-up and herded into a corral. Once inside the corral, they would receive their reward (conditioned response reinforcement). Another aspect is the association of the corral as a place of pleasure, where the animal receives sweet food (changing/training behavior), (Grant, 1964; Skinner, 1951).

Adhering to wildlife conditioning and enrichment techniques (Pizzutto, Scarpelli, Rossi, Chiozzotto, & Leschonski, 2013), we aimed to describe the strategies used during this study period to attract and herd free-ranging capybaras into confinement and assess the classical and operant conditioning paradigms as a viable aid for capturing capybaras. Moreover, considering the research objectives of the principal project, evaluating contraceptive effects, low-stress captures might have the least impact on the animal's hemostasis, especially on their reproductive physiology. Other positive aspects of concern include the need for smaller tranquilizer dosages, smoother procedures, quicker recovery, and more importantly, the ability to make frequent recaptures possible and safer.

Material and methods

Study area

The study was conducted year-round, during wet and dry seasons (December 2016 to January 2019) at the *Raia Olímpica*, a large man-made pool (Figure 2b) on the campus of University City, *Universidade de São Paulo*, Brazil. 23° 33' 21 S 46° 43' 14 W, altitude 722 m (WGS84 EGM96 Geoid. Google Earth Satellite Imagery, 2019). The pool area is approximately 247 500 m², surrounded by trees and extended grass areas, in the midst of one of the world's largest megalopolises (Figure 2). The Pinheiros river, the principal corridor for capybara migration, one of two rivers partially surrounding the City, is separated by an intercity highway. The city of São Paulo lies within the Atlantic Forest domains of the São Paulo State. Peak temperatures in the summer months (December - March) reach 30°C, with an average temperature of 25°C and median rainfall of 170 mm. In the winter (June - September), temperatures can fall to below 12°C, with an average temperature of 19°C and median rainfall of ≤ 50 mm (World Meteorological Organization, 2019).

Animals

In 2013, free-living capybaras invaded the pool area from the nearby Pinheiros river through a constructional breach in the water canalization system. A total number of two adults and five pups were reported to have entered, which grew to 40 animals within a four-year period then divided into two major groups and several satellite males (status at end of 2016).

Group I: 1 dominant male (original alpha male), 14 adults (including the original adult female), and 15 juveniles.

Group II: 1 dominant male (a direct descendant of the original alpha male and female). After growing into a sexually mature male, he was expelled by the alpha male, together with a second mature male (likely from the same litter). Over time, five adult females from group I joined the two males, forming group II.

Satellite males: distributed throughout the pool area; three sexually mature males that were driven out by the group I alpha male.

The polygynous society of capybaras represents a ridged linear hierarchy, where the alpha male agonistically protects his breeding rights. Nevertheless, subordinate females have been observed to leave the main group to reunite with satellite males, either temporarily or to form new groups. Likewise, subordinate males opportunistically attempt to mate with females from established groups (Herrera & Macdonald, 1993; Moreira et al., 2013).

Corral

For group confinement, we constructed a budget-friendly flexible corral (Figure 3a) using a scaffold frame (ea. element L: 150 x H: 100 cm). The leg extensions were forced into the ground and connected to one another using a metal wire (gauge 14). A continuous metal fence was then installed from the inside (1.5 x 45 m in circumference, $\pm 127 \text{ m}^2$), fixed to each scaffold frame using the same metal wire. The top portion of the fence was curved inward and maintained in position by tightening the upper portion of the fence every three to four m with metal wire. The aim of the fence's flexibility was to impede any push-throughs or possible jump or scaling escapes. Capture strategies for individual capybaras, namely satellite males, were twofold: (a) employ classical conditioning methods to lure the male into the corral, or (b) alternatively, use specially-constructed single-animal traps.



Figure 3. Corral & cage-traps. a) flexible corral design, b) single-trap, with removed doors for habilitation, c), depicting trapped capybara male (satellite).

Cage-traps

A single piece of 2 x 3 m (8 mm) steel net, with a 15 x 15 cm mesh, was folded to create a continuous U-shaped cage. The cage was placed over a pre-mounted steel frame and welded to all contact points. The structure was reinforced with steel rods. The U-shaped frame served as a track for the removable wood-plates, making the cage light enough to facilitate transport. The wooden front and back doors measured 100 x 115 x 1.2 cm, and the wooden floor measured 100 x 195 x 1.2 cm. The wood was made from a waterproof medium-density fiberboard (MDF) for external use. In addition, a wheel support was mounted onto each corner so that it could be rolled away. On location, these wheels were buried into the ground, assisting in keeping the cage in place. The trapping mechanism was based on two possible guillotine-door trigger concepts: by weight, pushing the floor down when entering, or by bait contact.

During habituation of the capybara satellite males to the traps, both sides of the doors remained open. Holes were placed on only one side of the wooden floor to allow the sugarcane to be placed upright (Figure 3b). When it was determined, via observation, that the animal was comfortable enough to enter the cage and feed, the cage was set for capture. The back-side was closed, the front door remained in the open position, the bait was set, and a simple nylon string was attached directly to the bait or to the floor to trigger the guillotine-door to slide shut (Figure 3c). The track also contained foam-cushion to minimize the noise of the door closing.

Behavioral observations

Thirty days prior to the first conditioning attempt (May 2017), behavioral observations were conducted, following descriptions by Altmann (1974), for three-hour periods during the day (either 07:00-10:00 or 14:00-17:00) or during the night (19:00-22:00). Observations were carried out at distances 1-100 m by direct observation or using visual magnifying aids and recording devices. The observations were conducted by a single investigator, studying the overall group dynamics; individual conduct; sleeping, resting, and eating periods; social interactions; and breeding behavior. The collected data served as a behavioral reference for observations after the contraceptive treatment.

Vocalization and body language as a stress indicator

To evaluate the success of our strategies in reducing capture and handling associated stress, we focused on direct behavioral observations (Skinner, 1938; Aguiar & Moro-Rios, 2009), such as conduct, body language, and vocalization. Capybaras have a broad spectrum of vocal communication (Sánchez, 2003; Barros, Tokumaru, Pedroza, & Nogueira, 2011; personal communication, 2017/2018). Interpretations were done by one observer and were based on vocal (V) and body language (BL) expressions for an individual's, and group's emotional state, discriminating between three principal states: (1) general well-being, curiosity, and positive excitement (V: high-pitched purring-clicking, often contagious to the rest of the group. BL: inquiring, probing, sniffing closing-in); (2) distress, fear, and irritation (V: squeaking, whistle, bark, teeth-chatter. BL: neck-hair stands up, muscle tremors, urinating, fleeing); (3) agonistic behavior (V: teeth chattering, bark. BL: neck-hair stands up, sham-attacks, real attacks, biting). We created several benchmarks for the purpose of measuring the progress of classical conditioning and behavioral training.

Classical conditioning

Initially, we used the concept of classical conditioning (or Pavlovian conditioning), taking advantage of unconditional stimuli (food) that was natural to capybaras, specifically sugarcane. Once attracted by the smell and voluntarily approaching the food source, referred to as the unconditional response (not based on behavior), we accompanied each food offering with click-like oral sound. This subsequently created a classical conditioning scenario by combining the unconditional response with a conditioned stimulus, which over a short period of time, turned into a conditioned response, providing the fundamental principles to expand the strategy to operant conditioning (or, Skinner Principle).

Conditioning goals: habituation to humans; voluntary approximation, hand-feeding; habituation to corals/traps; entering corrals/traps voluntarily (bait present); herding/entering corrals; and remaining calm during recovery. For each goal, the number/time of sessions needed for the first-time achievement was recorded.

Click-sound: starting with the first training session, a click-sound was used to achieve a conditional stimulus.

Positive experience: independent of scheduled training events, food was also placed in the corrals/traps to reinforce the association of the location with a pleasant experience.

Conditioning Sessions: fourteen days prior to the first capture event (June 2016), we initiated the conditioning process. Sessions lasted 30-60 min., three times a week, with continued behavioral observations conducted *ad libitum*.

Food for training reward/baiting

Capybaras are herbivorous, and although specific grass and aquatic plants are their staple food, they do have a great desire for sugary plants, especially sugarcane (lat. *Saccharum officinarum*) (Borges & Colares, 2007; Ferraz, 2007; Felix et al., 2014). Sugarcane was cut into approximately 2-foot-long pieces, and either hand-fed to the capybaras (Figure 4) or placed upright into the ground to attract the animals, which works much more effectively than leaving stalks uncut and flat on the floor (personal communication, 2017).



Figure 4. Conditioning/Training/Reinforcement w/ sugarcane; a) herding b) corral entry c) corral positive experience/reinforcement d) darted animal, while other members close by w/o agitation or agonistic behavior e) return of an treated animal, with members relocating towards the end of the corral, expressing no irritation or panic, and members outside the corral calmly grassing. (red arrows – sugarcane; yellow arrow – tranquilizer dart)

Calling/herding

Starting with the first contact, one observer emitted a click-sound to accompany each event, when approaching the animals or animals approached the observer, during food presentation/hand-feeding during herding (Figure 4a), and entering the corral (Figure 4b), with emphasis before and during food offerings. The click-sound was not used if food was not immediately associable, which was to avoid the negative effect of 'conditioning extinction', thus, possibly undoing the training attempts.

Positive reinforcements

Aside from direct training attempts, to eventually achieve an operant conditioning effect, efforts to complement the positive experiences with the event location were made. Food was placed periodically and independently from events, into the corral and traps, reinforcing their voluntarily entrance and pleasant experience during procedures and recovery (Figure 4c–e).

Chemical restraint

After the individual or group confinement, the target animal was anesthetized with a combination of ketamine and dexmedetomidine (9 mg kg^{-1} , Syntec, Brazil, and $5 \mu\text{g kg}^{-1}$, Zoetis, Brazil, respectively) which was administered intramuscularly into the hind leg musculature, using specialized darts and a CO_2 projector (X-Caliber, Pneu-Dart, Inc., Williamsport, Pa.).

Clinical procedures

After sedation, the animals were brought to our field clinic for biometric examination (weight, size, gender), ID marking, and health status evaluation. During different events, the procedures performed included administration of an immunocontraceptive, collection of semen, and in the final stage of the project, hemigonadectomy of the treated and control animals. After each procedure, the animal was returned to the corral, until fully recovered.

This study was approved by the Brazilian Ministry of Environment, SISBio: 54634-2 and *Universidade de São Paulo's* Ethics Committee for Use of Animals in Research, Ceuavet: 9553260816.

Results and discussion

In this work, our was to describe the consequences of developed capybara capture strategies that would permit frequent capture, without provoking severe discomfort to the animal in the form of stress or injury. Comparing this work with other literature is difficult because references relating to capybara capture methods are limited. One study describes a capture method using lassoing (Salas et al., 2004), which is the most dramatic form of live-capture besides culling and perhaps netting. With lassoing, three capture-dynamics are required: (1) the chase, (2) the actual capture by lasso, and (3) tying the legs. Although, within the report, capture-stress and risk of injury were not referred to, these actions must cause tremendous stress and a certain degree of injuries to the animals. However, considering the capture location and environment of Salas's study, this strategy was a necessary tool to capture target individuals. Several studies have been published in Portuguese that do mention corrals and baiting, even stating that it is the most appropriate method for capybara. They also provide some explanation of certain conditioning methods, types of bait used, and corral design, but do not elaborate on capture related animal-welfare, except death due to capture stress (Ferraz & Verdade, 2001; Rodrigues et al., 2017). One publication does describe, in length, corral design, trapping methods, efficiency, and types of bait, but does not mention anything about capture-related behavior and stress (Ferraz, Santos-Filho, Piffer, & Verdade, 2001). Several capybara-captures that used small-spaced, heavy-duty, closed-in, and steel-reinforced structures, with guillotine trap-doors, provoked capture-stress and panic behavior, causing injuries to the heads and legs of animals (personal communications, 2016).

Depending on the country and region, hunting-pressure can be considered an influential factor on behavioral and habitat changes, thereby influencing capture outcomes (Verdade, 1996; Moreira, Pinha, & Cunha, 2001; Moreira, 2013); however, within this specific area and study, this aspect is not relevant.

For the purpose of developing an alternative capture method, we initially employed the concept of classical conditioning (Grant, 1964; Owen & Amory, 2011) in association with specifically designed equipment. While assessing the theories of classical and operant conditioning, our goals were to attract and

herd free-ranging capybaras into confinement, making efforts to provide a positive experience, and site of pleasure, for the animals when voluntarily entering the corral, and during capture events, using sugarcane as the main bait and positive reinforcer. A further important aspect, considering the objectives of the principal research project on contraceptive effects, was the quality of the study results, influenced by low-stress and injury-free captures. We hypothesized that the capture strategies that we developed might have the least impact on the animals' hemostasis, especially on their reproductive endocrinology. In turn, this allowed for a more accurate comparison of the reproductive physiology under normal conditions compared with conditions under the influence of the contraceptive treatment. Other theoretical facets include the need for smaller tranquilizer dosages, smoother procedures, and quicker recovery.

We used two physical restraining models: (1) the corral design, which served as the principal, and preferred, method of capture, as this represents the method with the lowest impact on the animals; and (2) the single trap. This method was thought of as an alternative tool, to be used if target animals could not be conditioned, or captured within the corral, or were too distant from the corral, so herding would not be feasible.

Corral design

The large flex-corral withstood any escape attempt by the animals, usually achieved by probing the fence for weak spots (pushing the head into the fence at different locations). No scaling or jumping was observed. The fence's flexibility and the use of metal wire circumvented any breakage of the structure and prevented injuries to the animals. The corral's large oval shape allowed the trapped animals to relocate to different parts when humans entered to retrieve or return a sedated animal. We believe that this key design feature contributed to avoiding panic by providing enough 'safety-distance'. The prior habituation efforts, in conjunction with the 'pleasure' experience of finding reinforcing food in the corral and traps when entered voluntarily, contributed toward the calm capture, which was maintained throughout the duration of the animals' captivity.

Single-trap

After several cage-building trials, the final concept produced a cage/trap with a light-structure-design, allowing for easy relocation, as site adjustments were frequent. Although lightweight (33 kg), its structure was resistant enough to contain the trapped animal for prolonged periods of time. The trapping mechanism used was efficient in automatically trapping the animal.

Conditioned responses and behavioral targets

The first crucial task was to habituate the groups to our presence and for the animals to associate us and the click-sound with food. Surprisingly, very little time was needed to achieve this. The first session started with an initial distance of approximately 50 m, slowly moving toward the group while using the click-sound for the first time. Then the investigator stopped and remained in a position approximately 20 m away. The mood status of the animals was observed to be: (V) some animals barked; (BL) some animals got up, some were cautious or perhaps fearful, most of the group members turned and faced the direction of the water, and some juveniles dived into the water. Time invested at the location was approximately 20 min.

Voluntary approximation, hand-feeding

The second session, carried out on the following day, started from 50 m, the mood status of the animals was observed: (V), an initial two or three barks. The investigator approximation was done in intervals of 5 m, using a click-sound, until they remained approximately 10 m from the first members of the group. Sugarcane was forced upright into the ground, while also holding two pieces in the hand. The food was positioned up-wind to aid in the olfactory detection of the sugarcane. (BL), the animals closest to the observer got up into a sitting position, while the other animals remained as they were. Within five min., two individuals started to move toward the bait and carefully started to feed. Within a further three min., most members had gotten up and walked toward the sugarcane. The original members turned toward the investigator, hesitantly and cautiously starting to accept the food by hand. (V), animals made the singing-like high-pitched purring-clicking sound that is associated with positive excitement (personal communication, 2017). Throughout the entire session, the investigator emitted the click-sound intermittently. The goal of accepting human presence and hand-feeding was achieved in only two sessions and a total time of ≤ 50 min. was invested.

Habituation /entering the corral/trap

During the second session, the corral and trap were prepared with several pieces of sugarcane (Figure 4c). Some was placed at the entrance and some was distributed throughout the corral/trap. During the next visit on the following day, the sugarcane had been consumed, and there was evidence of capybara having visited, in the form of feces.

Herding attempts

Session three occurred two days after session two at approximately 09:00 and was the first capture event for procedures. The same investigator and two additional staff members, with sugarcane in hand, got close to the group. The distance between the group and the corral was approximately 220 m. While closing in, using the click-sound, the investigators stopped at a distance of approximately 10 m from the group. Within 2 min., the first members (apparently the same individuals from session two) came straight toward the food offerings. (V), on their way, they were observed to be emitting the purring-sound, and shortly after, three more members followed. The investigators used alternating 'stop' and 'go' to lure the capybaras toward the corral, using the bait as a tease and periodically allowing the animals to chew for a few seconds on the sugarcane, until the corral was reached (Figure 4a). Half-way through, some members started to stay behind, but they eventually reunited with the group. As the capybaras dictated the progress rhythm, the walk was slow; although, the goal of herding the animals over a lengthy distance was achieved.

Taking animals into the corral

After a 30 min. walk, the five individuals from the herding attempts would (BL) cautiously follow the investigator into the corral (the attraction of the bait seemed to be stronger than the uncertainty). (V), the animals would continue to emit the purring sound. The goal of entering the corral/first capture was achieved in one session, with the total time to capture being ≤ 20 min. (Figure 4b). We did recognize that the individuals initiating the approach, first to be hand-fed, and first to be herded and enter the corral, were not the dominant male or female (identified by body size and phenotypic characteristics). Another unexpected side effect, after taking the first five members into the corral, was that within the next 20 min. the entire group, including the dominant adults, had gathered around the corral, trying to get in, with multiple members emitting the purring sound. During this first procedure event, small pieces of sugarcane were given to the members periodically, inside and outside the corral, to maintain their interest and to sustain the overall tranquil atmosphere.

Remaining calm during procedures and recovery

During the anesthetic application and animal removal (Figure 4d) and after procedures, when returning sedated animals for recovery, the other group members relocated to the far-most side of the corral but remained tranquil (Figure 4e). (V), after the initial capture, no further purring-sounds were emitted. (BL), other than some probing at the fence, sitting down, or chewing sugarcane leftovers, no apparent fearful or irritated behaviors were observed, nor were any agonistic behaviors (toward other group members or staff). The goal of animals remaining calm during manipulation and recovery was achieved. The total time of the first event time (time of intervention) was approximately six hours.

After the first event, food was offered by hand-feeding, accompanied by the click-sound biweekly, during 30 min. reinforcement sessions. In addition, the corral was prepared with bait to maintain/reinforce the positive association with the location.

At each consecutive conditioning session, the response to the click-sound was almost immediate. In less than five min. (a fast response time for capybaras), most of the group members would approach the investigator to receive their food-treats, (V) making the purring-sound for the remainder of the feeding time. (BL), no stress-behavior was observed. Periodically, members and satellite males were observed inside the corrals, even without food present.

Over a period of 18 months, a total of 5 events were executed; each time the group members were successfully called and herded into the corral. Throughout all sessions and events, no agonistic or panic-like behavior was observed, except the occasional superficial attack by dominant animals toward subordinate members for the right to feed first.

Measuring behavior and stress

There are a myriad of techniques available to measure stress, which are mainly endocrinological and monitor stress-associated hormones or their metabolites. In addition, there are many well-defined ethological programs (DeNicola & Swihart, 1997; Romano et al., 2010; Copeland, 2015). However, for this project, because of logistics, resources available, and the irregular capture of free-ranging capybaras, the observational interpretation of vocalizations, behavior, and body language, was considered the only viable form of measurement. Although aware of potential investigator bias and the risk of subjective interpretation, driven by strong expectations (Tuytens et al., 2014), we believe that the results are clear and objective enough to avoid any misinterpretation.

All conditioning goals were achieved in relatively short time, such as habituation to human presence; voluntary approximation, allowing to be hand-fed; habituation to, and voluntarily entering, corrals/traps; and allowing to be herded over distances and taken into the corrals. Equally important, the aims to maintain a calm atmosphere when handling the animals in the corral, removing a sedated animal, or retuning the same animals for anesthetic recovery, with other members present, were all achieved. The conditioning method allowed the investigators to execute the selective capture of target animals at any time when needed, compared with random trapping. As there is no available literature on similar methods or experience with this species, it is not possible to discuss the time investment or quality of the conditioning.

Capybaras are inquisitive animals and, when there is no threat present, they are quite calm and approachable, especially if food is involved. How quickly they respond to offered food depends on the individual animal's character and experience, as well as the transient group dynamics. As soon as one member breaks the contact barrier, the rest will follow shortly, suggesting a learning behavior by imitation. The capybaras quickly learned to associate the tongue-click sound with food, and after only a few repetitions, the approaching vehicle and individual staff members were associated with food and greeted with excitement.

Great caution is warranted to not feed excessive amounts of sugarcane to capybaras, as this may lead to intensive fermentation, potentially provoking tympany. In addition, working with hand-fed bait, a calm situation can quickly turn into a light feeding-frenzy when the animals get too close to each other, leading to fights of feeding-dominance.

Based on our behavioral observations, the capture strategies employed in this study appeared to provide several positive effects. Using the concept of classical conditioning was successful, with animals rapidly associating the click-sound with food offerings and approaching the investigator to be hand-fed. Animals could easily be herded into the corral while maintaining a calm demeanor throughout all capture events. These results demonstrate that the applied conditioning methods for capybaras are useful in facilitating frequent capture needs.

Conclusion

There is no 'one-strategy fits all', and each situation and species demand its own capture strategies. Nevertheless, for the purpose of frequent capture, we propose that for capybaras, where time and logistics permit, prior conditioning can be a beneficial strategy in preserving the capybaras' overall welfare while facilitating the execution of frequent capture and procedures.

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We declare that this article is original and has not been submitted for publication in any other national or international journal, either in whole or in part.

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