The Use of Operant Conditioning (Clicker Training) of Göttingen Minipigs for Topical Safety Studies

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ABSTRACT

The Gottingen minipig is a widely used large animal model in pre-clinical drug safety studies. Using standard restraint techniques, applying topical compounds to the backs of minipigs can be challenging because minipigs are often fractious. Operant conditioning is used with various species for general care and research, and has been used with pigs in discrimination research, animal welfare research, cognitive science, and animal science. Our goal was to use this method to train minipigs to voluntarily cooperate in experimental procedures, thereby enhancing animal welfare and providing enrichment. Technical colleagues were trained in the theory and practice of operant conditioning (clicker training). By pairing the sound of a click with food, animals learned that they would be rewarded for performing certain behaviors. Training lasted approximately fifteen minutes per day per pig over a three-week period. Defined protocols were used to train naïve minipigs to approach handlers, follow a target stick, walk onto a scale for weighing, and stand still for physical examinations, electrocardiography, and dermal dosing. Training was conducted with minimal physical handling using only positive reinforcement. These techniques have become standard practice in our facility, and have created a positive environment where animals voluntarily present desired behaviors. Training minipigs has resulted in significant reductions in a) injuries to animals and staff, b) stress levels of animals and staff and, c) the number of staff required for procedures. This training enriches the animals' environment and their interactions with handlers. Operant conditioning is a cost-effective way to provide a positive environment for animals and staff, and has ergonomic and economic benefits. We would like to report the first use of operant conditioning on minipigs in preclinical safety studies to diminish the stress of performing dermal studies, and encourage adoption of operant conditioning techniques in other research studies on large animals.

INTRODUCTION

The Göttingen Minipig as a Model for Preclinical Dermatology Studies

Advantages

- Minipigs are considered the best animal model for percutaneous absorption because their skin is morphologically and functionally similar to human skin
  - Thickness of integument
  - Haircoat
  - Stratum corneum lipid composition

Disadvantages

- Conducting the study is very labor intensive
  - Four technicians are required for dermal dosing
  - Six technicians are required for blood collection
- There are concerns for employee safety
  - Physical strength is required to handle, dose and perform procedures on the animals
  - Animals grow quickly during a month-long study
  - Mature males can occasionally be aggressive

Initial Attempts to Improve Minipig Behavior

Acclimation Program 1
Week 1: In-cage socialization
Week 2: Handling, restraint in sling
Results: Improved behavior, but minipigs remained difficult to handle

Acclimation Program 2
Prior to arrival – socialization and handling by vendor (2 weeks)
After arrival – 4 weeks in-house acclimation
Week 1 – hand feeding
Week 2 – handling
Week 3 – sling restraint
Week 4 – dorsal recumbency
Results: Initial improvement in behavior, followed by regression


Typical minipig pen


Göttingen Minipig

HYPOTHESIS

After networking with colleagues in our department, the idea of using operant conditioning on minipigs was explored. Operant conditioning is used in a wide variety of settings (e.g., research laboratories, zoological parks, and veterinary clinics) with many animal species, both wild and domestic.

We hypothesized that minipigs can be trained using operant conditioning to be cooperative and willing participants in studies involving dermal dosing, electrocardiogram recording, and physical examination, including body weight determination.

Scientists, technicians, and veterinarians collaborated and formulated a clicker training program for the minipigs, which was established in June 2005. In September 2005, Robert Bailey, an animal behaviorist specializing in operant conditioning, conducted an educational seminar and clicker training workshop at our site. Further refinements were made to our clicker training program. This work was performed in an AAALAC-International-accredited facility and all experimental procedures were approved by the PGRD-Ann Arbor IACUC.

THEORY

Classical conditioning is used to "charge" the clicker (pair a "click" sound with the presentation of food). In operant conditioning the clicker then functions as a bridge between a behavior and a food reward, and marks the precise moment the animal is performing the desired behavior. Behaviors are trained by shaping using very small steps; the animal must master one step before learning the next.

Classical Conditioning

- Control of a physiologic response with a neutral stimulus (e.g. Pavlov's experiments)
  - Dog sees food and salivates (involuntary response)
  - Sound paired with food elicits salivation
- Acclimation – animals must be calm before they can learn

Operant Conditioning

- Active learning process where the animal offers behaviors
- A positive consequence (food) increases the probability of the animal repeating the behavior

APPLICATION

Acclimation Program 3
Prior to arrival – socialization by vendor (2 weeks)
- Handling and hand feeding
- Acclimation to airline crates
- Animals are shipped by truck 2 per crate

Upon receipt
Transport to animal room in shipping crates; procedures are performed before animals are place in pens:
- Physical exam performed by a veterinarian
- Body weight and temperature recorded
- Microchip implant inserted

Minipigs are individually housed in pens containing a half airline crate, and food and water are provided

Day 4
- Crate removed
- Clicker training commences as outlined in Table 1

The training program starts two to three weeks prior to study initiation. Training is conducted every business day for approximately 15 minutes/pig/day. The trainer grades each animal's proficiency/fluency for each step of each behavior and the results are documented. Progression to the next step or behavior is dependent upon each individual animal's competence (Table 2).

TOOLS

Plastic spoons were used as target sticks. Airline crates equipped with mats, ladles, and plastic disks were used to hold the animal for procedures, and physical restraint was not required. Animals received only their allotted daily rations; no extra food was needed.


Target Stick


Crate with Installed Target

TABLE 1: TRAINING SCHEDULE

Sunday	*Monday*	*Tuesday*	*Wednesday*	*Thursday*	*Friday*	*Saturday*
				Animal Arrival	Acclimation No Training	No Training
No Training	Training Begins • Charge Clicker	• Charge Clicker • Target Stick	• Charge Clicker • Target Stick	• Target Stick • Crate Train	• Target Stick • Crate Train	No Training
No Training	• Target Stick • Crate Train • Fixed Target in Crate	• Target Stick • Fixed Target in Crate • Crate Transfer • Dermal Dose Training	• Target Stick • Fixed Target in Crate • Crate Transfer • Dermal Dose Training	• Target Stick • Fixed Target in Crate • Crate Transfer • Dermal Dose Training	• Target Stick • Fixed Target in Crate • Crate Transfer • Dermal Dose Training	No Training
No Training	Study Initiation	Behaviors reinforced during course of study • Dosing (≈ 10 minutes) • ECG's • Body Weight Collection				

TABLE 2: TRAINING PROTOCOLS

Charge Clicker	• Click and put pellets in pan (Click and treat; C&T); repeat • Continue until animal is confident and looks for the food after the click • Move pan around to acclimate animal to being in different areas relative to the trainer
Target Stick	• Introduce target stick, C&T for interest in target • C&T for closer proximity to the target • C&T for touching target accurately • C&T for touching target longer
Crate Training	• Use target stick to lure minipig into crate, feed from the pan • C&T for touching target stick • Bring target stick into crate at ~1 foot intervals. Do not advance until minipig is confident at each step • As minipig approaches the end of the crate, place the food into the installed ladle rather than pan • Guide the minipig out of the crate with target stick when ready, feed from the pan outside of the crate
Fixed Target in Crate	• When minipig is confident standing in the crate touching target stick, place the target stick over the installed target • Move the target stick away and C&T for touching the installed target
Crate Transfer	• Set up the crate outside the pen • Use target stick to guide minipig into crate, C&T for touching stick • C&T for touching stationary target • Guide out of crate with target stick • C&T when back in pen
Dermal Dose Training (two trainers required)	• C&T the minipig for touching the target while the trainer does the following: – Start with hand resting on crate – Move the hand into air over the animal – Put hand over animals back – Let hand rest on animals back – Gently stroke animals back – Rub animals back – Acclimate animal to dosing rod – Acclimate animal to dosing procedure using water


Dermal Dosing


Body Weight Determination

RESULTS

We have now successfully trained a total of 94 minipigs for use in 10 dermatology safety studies. After a training period of 12-15 days, behaviors are strong enough to maintain excellent behavior through a thirty-day study period. Using this program, only two technicians are required for dosing, ECG collection, body weight determination and physical examination. Two or three technicians are required for blood collection.

CONCLUSIONS

Using this program of operant conditioning, we have

- Increased animal enrichment
- Developed a more cooperative and willing subject
- Increased safety for animals in study procedures
- Increased accuracy in dosing animals topically
- Increased efficiency and productivity
- Greatly reduced stress to animals and staff
- Exceeded IACUC and other guidelines for animal care and enrichment
- Provided an opportunity for staff career development by learning this training method
- Minimized the number of staff required for study procedures
- Reduced cost
- Reduced the chance of injuries (i.e. bites, kicks, bruises)

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