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Case Study: Training a Chimpanzee (Pan troglodytes) to Use a Nebulizer to Aid the Treatment of Airsacculitis

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Bacterial airsacculitis has been reported in a variety of nonhuman primates, and is widely treated using a combination of surgery and oral antibiotics. This case study details an alternative method of administering antibiotics (via the use of a nebulizer) when the chimpanzee subject developed resistance to all available oral preparations. Training the chimpanzee to use the nebulizer was performed using positive reinforcement techniques (PRT). It took a total of 89 sessions (<7 hr 25 min) to train the chimpanzee to use the nebulizer. The airsacculitis infection was treated using colistin in the nebulizer twice a day for 9 days. Out of 18 potential treatment sessions, full doses were administered on 13 occasions. The final dose of colistin was given via slow brachial intravenous injection under general anesthesia. The infection was successfully treated with colistin. Although there was a training time investment involved, it was felt to be outweighed by the success of the treatment. Also in the likelihood of the infection re-occurring at a later date, the now learnt behavior of using a nebulizer means that future treatment should now be considerably quicker. This is another example of how PRT is a useful tool in the successful welfare and management of captive animals.


Keywords: positive reinforcement; PRT; desensitization; primates

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INTRODUCTION

Bacterial infections of the air sac (bacterial airsacculitis) have been reported in a variety of nonhuman primates, including owl monkeys [Giles et al., 1974], orang-utans [Cambre et al., 1980; Lawson et al., 2006], and chimpanzees [Hill et al., 2001; Strobert and Swenson, 1979]. Complications in airsacculitis can be fatal [Cambre et al., 1980], yet often a distended air sac, upper respiratory tract infection, or halitosis may be the only apparent clinical signs [Hill et al., 2001; Lawson et al., 2006; Strobert and Swenson, 1979].

Bacterial airsacculitis in chimpanzees and other nonhuman primates has successfully been managed by a combination of surgery to allow drainage of the pus and antibiotic therapy [Hill et al., 2001; Lawson et al., 2006; Strobert and Swenson, 1979]. As described by Hill et al. [2001], suitable oral antibiotics are an effective way of treating the individual after surgery, as the animal can remain in its social group, but what happens when the bacterial infection develops a resistance to all available oral antibiotics?

The subject of this study was Emma, a 27-year-old female captive born chimpanzee, housed at the Royal Zoological Society of Scotland’s (RZSS) Edinburgh Zoo site. She had a 10-year history of chronic air sac enlargement, which had been investigated under numerous anesthesias. In January 2008, keepers noticed that her air sac had become larger and appeared to have changed in consistency. In April 2008, surgery was performed under general anesthesia. Through an incision into her air sac, a large amount of pus was drained, and a single stoma was created to allow for further drainage and future cleaning. *Pseudomonas aeruginosa* was isolated from the samples taken at the time of surgery, and was initially sensitive to ciprofloxacin (an orally administered antibiotic). During treatment with ciprofloxacin, subsequent samples from the air sac still revealed a *P. aeruginosa* infection, but on these occasions, it was multi-resistant to numerous antibiotics. The results did show sensitivity to amikacin, tobramycin, gentamicin, and colistin, which are not available as oral preparations. Of these drugs, colistin (a drug developed for human use, Colomycin; Forrest Laboratories UK Ltd, UK) can be administered via nebulization as well as intravenously.

It has been shown that chimpanzees can be successfully trained via positive reinforcement techniques (PRT) to successfully cooperate in a variety of veterinary procedures: obtaining urine [Lambeth et al., 2000; Laule et al., 1996; Stone et al., 1994; Whittaker et al., 2001] or blood samples [Coleman et al., 2008; Laule et al., 1996; Whittaker et al., 2001], presenting to receive injections [Coleman et al., 2008; Lambeth et al., 2006; Laule et al., 1996; Shapiro et al., 2003; Viede et al., 2005; Whittaker et al., 2001], and learning body examination behaviors [Whittaker et al., 2001]. The chimpanzees at RZSS Edinburgh Zoo were already trained by their keepers via PRT in order to assist with various husbandry and veterinary needs. This included stationing, target training, and presentation of named body parts. The chimpanzees were trained using operant conditioning, with the keepers using clickers to act as a bridge between the performance of the desired behavior on cue and an up-and-coming reward.

Emma’s specific training plan had been designed to include the presentation of her chest to the bars of the cage on cue (by verbal command and hand signal). This allowed the keepers to manipulate her air sac, clean the stoma and also to take a bacterial swab from the stoma for routine testing. The keepers’ previous attempts to
train her to cooperate with injections were not proving to be very productive. Emma had recently received numerous injections via dart (for husbandry and veterinary requirements). This understandably seemed to have had a negative effect on her choice to participate in PRT injection training. Based on this information and veterinary advice, it was felt that her existing chest inspection programme could be expanded to allow nebulization through the stoma.

Therefore, there were two training and veterinary objectives:

1. To train Emma to use a nebulizer.
2. To treat Emma against the *P. aeruginosa* infection with colistin via a nebulizer.

The aim of this article is to describe how it was possible to train Emma to use a nebulizer via PRT and how successful this method was in administering the antibiotics required as part of her airsacculitis management plan.

**MATERIALS AND METHODS**

The creation of the stoma in Emma’s air sac meant that the medication could potentially be pumped into her air sac from here. This would negate the need to train Emma to put her face into a nebulizer mask. A portable compressor nebulizer system was used (Clenny Aerosol, Medel CE0123). This was chosen because it was relatively small and quiet when switched on. It was adapted by detaching the mask from the tube connected to the machine and for final administration the tube tip needed to be modified to maximize airflow into the stoma. Being that the tube tip was designed for insertion into a nebulizer mask as oppose to a chimpanzee air sac stoma, it was found that air (and potential medication) escaped before entering the stoma. Therefore, the latter part of the tube was removed to prevent this.

**Training Emma to Use a Nebulizer**

This period of training took place at the Budongo Trail exhibit, RZSS Edinburgh Zoo from July 15–October 17, 2008. A step-by-step PRT program was designed to help to train Emma to use the nebulizer. Because Emma was already trained to have a bacteriology swab inserted into her air sac stoma, this was used as the starting point for the programme. The keepers at Budongo Trail have themselves been trained in PRT, and as they have the best relationship with Emma, they were appointed to do the training.

Each training session lasted no longer than 5 min, and was performed at most, once a day. This schedule was picked because it fitted in well with the daily husbandry routine and also this level of training with the chimpanzees had previously yielded good results in teaching them other behaviors. It has since been shown in rhesus macaques that this level of training was more efficient than either twice daily or three times a week [Fernstrom et al., 2009]. The session was only ended before this time if Emma left (for whatever reason) and did not return within the allocated 5 min, or if she was issued with three separate time-outs per session. A time-out (where the keeper took a step back and ignored Emma for 30 sec) was only used if Emma threatened the keepers, tried to scratch them or grabbed the nebulizer tube.

A clicker was used as a bridge to signal to Emma that she had performed the behavior correctly and for long enough. The sound of the clicker indicated to Emma that she would receive a reward (a drink of weak sugar-free juice or a piece of dried...
fig, all out of her daily ration), therefore reinforcing the behavior. The ultimate aim was achieved by shaping the entire behavior and breaking it down into a series of attainable stages:

1. Air sac cleaned and swabbed with nebulizer machine in view of Emma, but off.
2. Insert nebulizer tube into air sac stoma with machine off
   (a) Insert tube into air sac stoma briefly (<2 sec).
   (b) Insert tube into air sac stoma, and increase time inserted until 10 sec reached.
3. Insert nebulizer tube (with restricted air flow) into air sac stoma, with machine on
   (a) Insert tube into air sac stoma briefly (<2 sec).
   (b) Insert tube into air sac stoma, and increase time inserted until 10 sec reached.
4. Modified nebulizer tube inserted (with maximized airflow) into air sac stoma, with machine on
   (a) Insert tube into air sac stoma briefly (<2 sec).
   (b) Insert tube into air sac stoma, and increase time inserted until 10 sec reached.

At the start of each attempt to train Emma to use the nebulizer, she was given the cue to present and hold her air sac to the mesh. Because Emma was never separated from the rest of her cagemates, if other individuals were present, they were positively reinforced by another keeper for not interfering with her training. At no point throughout the training process did Emma receive medication, even when the machine was switched on.

Ideally only one keeper would shape the entire behavior, to keep consistency. However, due to staffing levels and other commitments, plus the pressing need for Emma to be trained to use the nebulizer in order to receive her medication, this would mean being able to commit to only one session every 2 days (as opposed to one session daily). Therefore, two keepers were allocated to shape the process (the primary trainers), and to help ensure consistency, a detailed diary and regular verbal communication occurred between the two keepers. If for any reason both these keepers were absent, a third keeper took the training session, but only trained up to stage 1 in the program (once it had become a learnt behavior). These and any other sessions that only trained learnt behaviors will be subsequently referred to as maintenance training (MT) sessions.

During the shaping process, Emma had to perform the required behavior stage consistently throughout two consecutive training sessions before the keepers would move onto the next stage in the program. Ideally each stage may have been reinforced more, to ensure that it was a part of her repertoire, but the need to administer the medication meant fewer sessions were used in the reinforcement process. The entire behavior (i.e. up to stage 4b) was deemed to be learnt reliably, once she had consistently performed the entire behavior for 9 out of 10 consecutive sessions. The third trainer could then train this learnt behavior.

**Treatment of *P. aeruginosa* With Colistin via Nebulizer**

The treatment period took place between October 22 and October 30, 2008. It was possible that Emma would experience a slightly different sensation when using colistin through the nebulizer, as opposed to just air, but it was presumed that this slight difference should not affect her behavior greatly if she was already fully desensitized to the nebulizer. As advised by the vets, colistin powder at 500,000 units per vial was mixed with 2 ml of saline and nebulized via the stoma. The aim was to
insert the nebulizer tube for 10-sec periods during two training sessions per day, culminating in at least 2 min of nebulization per day for 9 days. The final dose of colistin was given via slow brachial intravenous injection (500,000 units per vial diluted in 5 ml of saline), during a planned general anesthesia 10 days after the nebulizing treatment had finished. The anesthesia was needed to test her response to the nebulizing treatment. Since intravenous administration is the more commonly used route for this drug, the opportunity was taken to administer the final dose parenterally.

RESULTS

Training Emma to Use a Nebulizer

As can be seen in Table 1, it took 89 sessions (<7 hr 25 min) in total, spread over 3 months to achieve the aim of desensitizing Emma to the use of a nebulizer. This total number of sessions included any MT sessions that took place during this period. There were 19 MT sessions in this total period, meaning that it took 70 sessions (<5 hr 50 min) of actual shaping in the desensitization process.

Stage 1 (cleaning and swabbing Emma’s air sac with the nebulizer machine switched off) was the quickest for Emma to learn, having performed so in the minimum amount of 2 of the 89 sessions (2.2%). Stage 4 (air pumped into the stoma with the nebulizer tube tip cut off) took the longest amount of time to train, accounting for 43 of the 89 sessions, nearly half the total (48.3%). Stage 2 took longer (26 of 89 sessions) to train than stage 3 (18 of 89 sessions), and the latter part of stages 2–4 (parts b) took longer to train than their counterparts (parts a).

From time to time, Emma flinched when her air sac stoma was touched, and sometimes pulled away from her training position (Fig. 1). It is reasonable to presume that this meant touching her stoma gave her discomfort; at these times it was assumed that the stoma was more sensitive, although it was not proven so. To minimize any discomfort when training, the keepers were often unable to progress at these times. Emma’s stoma was recorded as sensitive in 24.7% of the total training sessions, with just over half (54.5%) of these occurring during the training of stage 2 (inserting the nebulizer tube with the machine switched off). The remainder of these occasions occurred in stage 4 of her training.

<table>
<thead>
<tr>
<th>Stage</th>
<th>No. sessions (excluding MT)</th>
<th>No. sessions stoma was sensitive</th>
<th>No. sessions Emma terminated early</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>2 (2)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2a</td>
<td>4 (4)</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>2b</td>
<td>22 (16)</td>
<td>11</td>
<td>4</td>
</tr>
<tr>
<td>3a</td>
<td>6 (4)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>3b</td>
<td>12 (8)</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>4a</td>
<td>4 (3)</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>4b</td>
<td>39 (33)</td>
<td>10</td>
<td>2</td>
</tr>
<tr>
<td>Total</td>
<td>89 (70)</td>
<td>22</td>
<td>9</td>
</tr>
</tbody>
</table>

Zoo Biology
Emma always had the option to terminate the session by leaving early. She only did this in 9 out of the 89 sessions (10.1%). The majority of these early terminations occurred in stage 2 of the training.

Treatment of *P. aeruginosa* With Colistin via Nebulizer

Out of a maximum 18 potential treatment sessions, she successfully received a minimum of 1-min culminated nebulization with colistin on 13 occasions. On October 23 and 25, only one session per day was conducted due to staff absences. In the remaining three sessions, she did not receive a full minute of nebulization due to her air sac being particularly sensitive at these times. This made nebulization virtually impossible without causing pain to Emma, which was important to avoid for both her short-term and long-term welfare if she could become less likely to participate in future sessions.

The training and treatment took in total 117 sessions (<9 hr 45 min), including a period of MT in between the desensitization training and treatment period. During this time, other cage mates only attempted to interfere with Emma’s training on six occasions, by only 2 out of the remaining 10 individual chimpanzees.

Microbiology swabs were taken from inside the air sac via the stoma at the start and at the end of treatment. This allowed the response to the treatment regime to be monitored. Swabs were also taken regularly after the completion of treatment to monitor the air sac long term.

DISCUSSION

Successful treatment plans for airsacculitis in nonhuman primates have often incorporated surgery and antibiotic therapy [Hill et al., 2001; Lawson et al., 2006; Strobert and Swenson, 1979], and in Emma’s case, this was no different. However, the complication arose in this case study when Emma developed a multi-resistant infection to oral antibiotics. The training of Emma to use a nebulizer was just one aspect in the long-term management of her air sac infection, yet shows again how valuable PRT can be within the realms of captive animal management.
Training Emma to use the nebulizer could initially have been seen as a fairly daunting task, but as with training any complex behavior, once broken down into small attainable steps, it became a very achievable mission. 117 sessions (9 hr 45 min) spread over 3½ months enabled her to be trained and treated. This figure included all the MT sessions. It is impossible to say whether or not the MT slowed down the entire training process; without them, more shaping sessions may have been required. Although they were not an integral part of the training program (having been performed mostly to cover staff absences), they probably would have provided valuable behavioral reinforcement.

Some of these steps took longer to achieve than others. Although the stages appeared fairly uniform on paper, the sensations exposed to Emma in the final stages of training could have increased in terms of novelty and perhaps discomfort. The final stage (where the modified tube was used) meant that her air sac expanded in size more rapidly than before. It could be interpreted that this stage was more unpleasant to her than the previous stages, judging by the time it took her to become desensitized to this sensation. Even discounting the 10 sessions where the stoma was assumed to be sensitive, stage 4 still took the longest to achieve.

A surprising result was that stage 2 (inserting the nebulizer tube with the machine switched off) took longer to achieve than stage 3 (inserting the tube with the machine switched on). It was felt that the sensation of air inflating her air sac would be a novel experience, and so possibly would take longer to train. However, 12 out of 26 of these stage 2 sessions occurred when her air sac was assumed to be sensitive and Emma chose to leave 4 of these sessions early, perhaps an indication that this potential sensitivity played a role in slower progress.

Stages 2–4 were broken down further into two parts (a and b). In each of these stages, part (a) consisted of brief nebulizer tube insertions (<2 sec), whereas part (b) increased these insertions up to 10 sec. This insertion time difference in the two parts probably accounted for the higher time required to train part (b) than (a).

Training Emma to voluntarily use a nebulizer meant that her infection could reliably be monitored and treated. It also increases the variety of antibiotics she could potentially receive (including those developed for human use), which is beneficial when dealing with an infection that is multi-resistant. Once the use of the nebulizer became a learnt behavior, it meant that other keepers could then train her in this procedure. It also opens up the opportunity for this method of management to be used by other institutions. Understandably, this was a very unique situation, and part of the success of this treatment regimen was down to Emma’s personality and willingness to participate in PRT. Therefore, it is important to recognize that individuals do show variances when trained by PRT [Coleman et al., 2008; Desmond and Laule, 1994; Prescott et al., 2005; Prescott and Buchanan-Smith, 2003; Shapiro et al., 2003; Videan et al., 2005].

In this study, out of the 117 sessions recorded, interference from other members of the group only occurred on 6 separate occasions, and this was restricted to two individuals. If this became a persistent problem, these individuals could be positively reinforced not to interfere with Emma’s nebulization. The use of PRT meant that Emma could be treated without having to separate her from the rest of her cagemates, which for a social species like the chimpanzee, is an important factor. Removing an individual can cause stress not only to the subject, but also to those left behind [Shapiro et al., 2003]. A reduction in stress can aid the psychological
CONCLUSIONS

1. Training a chimpanzee by PRT to use a nebulizer can aid the successful short- and long-term management in the treatment of airsacculitis.

2. PRT (and therefore the chimpanzee’s voluntary cooperation) helped staff to maintain high standards of animal care and welfare.

3. Although an initial time investment was required, this was felt to be far outweighed by the health benefits gained, both short and long term.

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