

## **SOCIALIZATION OF DUTCH BELTED RABBITS FOR USE IN RESEARCH: PRELIMINARY OUTCOMES**

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### **ABSTRACT**

Dutch belted rabbits are commonly used as research animals due to their small size compared to New Zealand White rabbits. As a generalization, Dutch belted rabbits tend to be approximately 2/3 smaller than New Zealand Whites in our colonies. In the laboratory, this translates to the utilization of less test article and easier manipulations of the animals with fewer worker injuries. However, in the United States, it has been reported that commercially raised black Dutch belted rabbits are more aggressive towards their human handlers and less adaptive to environmental change than their New Zealand White counterparts.

An intensive socialization program was initiated in a colony of black Dutch belted rabbits raised exclusively for research. This program includes adaptation to common animal handling procedures, environmental enrichment devices, and other materials encountered in the laboratory. This program encompasses handling pre-and post-weaning and prepares the animals for use in research with less overall aggression to human caretakers and less stress to novel environmental stimuli. Success of the program has been measured in terms of a decrease in the number of complaints received about animal behavior once the animals have arrived at the research facility.

**Key words:** Dutch belted rabbits, Rabbit behavior, Research laboratory, Socialization, Environmental enrichment.

### **INTRODUCTION**

In response to reports of aggression towards human caretakers and excessive stress in novel situations in a colony of Dutch belted rabbits raised exclusively for research, an intensive socialization program was initiated. This program includes adaptation to common animal handling procedures such as physical exams, environmental enrichment devices such as toys and food treats, and other objects encountered in the laboratory such as transport containers. This program encompasses handling pre-and post-weaning and prepares the animals for use in research with less overall aggression to human caretakers and less stress to novel environmental stimuli.

Socialization is begun at three days after kindling and continues until either the animals are shipped to a research laboratory or retained for future breeding. This program thus encompasses rabbit handling during pre-weaning which can help the animals become less timid and display less negative behaviors towards humans as adults (Kersten *et al.*, 1989). Additionally, these adult animals will display fewer fear behaviors which are viewed negatively by caretakers and more exploratory behaviors which are viewed positively by caretakers (Kersten *et al.*, 1989; Jezierski and Konecka, 1996). We chose to initiate our program at 3 days of age to take advantage of a sensitive development period of the pups during the first week of life and promote observations of animal health (Kersten *et al.*, 1989; Jezierski and Konecka, 1996; Csatádi *et al.*, 2005; Csatádi *et al.*, 2007). The goal of fostering interactions with novel enrichment devices and other items builds on the knowledge that rabbit pups may show a reduced fear towards new items when they have been exposed to such items for short periods of time (Pongrácz *et al.*, 2001).

## MATERIALS AND METHODS

### Animals

The animals used are black Dutch belted rabbits aged 3 days to 12 weeks. The animals are raised in a closed colony at a breeding facility accredited by the Association for Assessment and Accreditation of Laboratory Animal Care, International (AAALAC, International). Animals are housed in three tiered stainless steel caging. Water is provided on an *ad libitum* basis. Feed (Purina, Richmond, Indiana, USA) is provided once a day in measured amounts based on age and body weight. The facility maintains at least 10 air changes per hour of fresh air and a complete heating/ventilation/air conditioning (HVAC) system provides for temperature control at 65-75°F and relative humidity control at 30-70%. The colony is free from the following pathogens and conditions: *Pasteurella multocida*, *Pasteurella pneumotropica*, *Salmonella*, *ectoparasites*, *Eimeria stiedae*, *Treponema cuniculi*, *Clostridium piliforme*, *Cilia Associated Respiratory (CAR) Bacillus*, *Encephalitozoon cuniculi*, *Toxoplasma gondii*, *Passalurus ambiguus*, and *dermatophytosis*.

### Objects Utilized

- Stainless steel rattlers (Bio-Serv, Frenchtown, NJ, USA)
- Wiffle balls (Various manufactures, USA)
- Dumbbells (Bio-Serv, Frenchtown, NJ, USA)
- Plastic balls with stainless steel washers (“Jingle Balls”, Bio-Serv, Frenchtown, NJ, USA)
- Fruit flavored treats (“Fruity-Gems”, Bio-Serv, Frenchtown, NJ, USA)
- Latex gloves (Various manufacturers, USA)
- Plastic restrainers (Tecniplast, Exton, PA, USA)
- Plastic transport carts (Rubbermaid, USA)
- Scale (American Weight Systems, Mountville, PA, USA)
- Transport boxes (Timbar, Hanover/New Oxford, PA, USA)

### Experimental Design

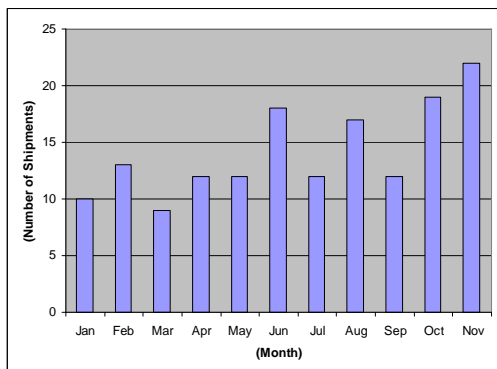
1. The first interactions with humans occur at 3 days post-kindling when the first nest box checks are performed. Checks continue twice a week until 28 days post-kindling for a total of eight checks prior to 28 days of age. The pups are handled for a few seconds to a few minutes at each check to assure animal health and begin socializing with humans. Checks occur in the very early morning (approximately 5 to 6 AM). In this intensive breeding operation, nursing typically does not occur during the daytime (4 AM to 4 PM with artificial lighting) but instead generally occurs during the nighttime (4 PM to 4 AM with artificial lighting). It is thought that nursing occurs in the morning hours prior to daytime. Thus, the pup checks were not associated with a specific time after nursing.
2. At 28 days post-kindling, the nest box is pulled from the cage. Pups are weaned at 5 weeks of age.
3. Immediately after weaning, the rabbits are placed in group housed cages for approximately 2 weeks.
4. At 7 weeks of age, the rabbits are paired with a partner for two weeks. Intensive socialization begins at this stage. Once a week, rabbits are brought out onto the top surface of a plastic transport cart, similar to what is done in research facilities. The rabbits are petted and their teeth, eyes, and toes are examined. Their nails are trimmed and special attention is given to manipulations of the abdomen. On the cart, novel objects are placed and the rabbit is given 10 to 15 minutes to explore them. The objects include fruit treats, enrichment devices, and latex gloves. The rabbits are again petted during this exploratory time.
5. At 9 weeks of age, the rabbits are typically identified as to which research facility they will be going to. Approximately equal numbers of male and female rabbits were pulled for orders and socialized, although orders may consist of males only, females only, or a mix (usually 50%

male and 50% female if a mix). Four times before they are shipped, the rabbits undergo the following socialization program:

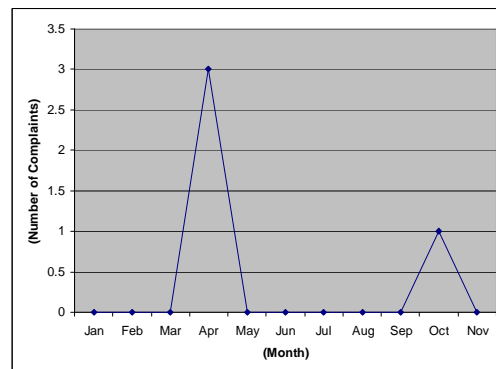
- a. Physical exams
  - b. Acclimation to a plastic restrainer for 15 to 120 (maximum) minutes followed by a physical exam. If the animals have difficulty adapting to the restrainer, they are held by caretakers and calmed down before repeat attempts.
  - c. Placement in a transport container for 30 to 60 minutes with the lid off or on followed by a physical exam.
6. Does identified for future breeding are socialized on the surface of the transport care three more times before kindling their first litter. Environmental enrichment devices are placed in their cages up until the time of breeding.

## RESULTS AND DISCUSSION

The socialization program was initiated in May of 2007. For 2007, there was a decrease in the number of complaints per month (0.75 for January through April and 0.14 for May through November) since the initiation of the socialization program despite the increase in number of shipments (11 per month for January through April and 16 per month for May through November) (Figure 1 and 2). All complaints received through April were for male Dutch Belted rabbits aged 5 to 7 months. The complaints covered approximately 150 rabbits out of multiple shipments, representing 16.2% of the total number of Dutch Belted rabbits sold during that timeframe. The single complaint received May through November was for one order of four female rabbits aged 5 months. This represented 0.2% of the total number of Dutch Belted rabbits sold during that timeframe. A survey for clients to complete and return back is sent with every order. Additionally, we maintain a rigorous complaint system whereby clients are able to call in their concerns and have them logged into a complaint monitoring system. Clients are very proactive about sending us concerns and issues regarding shipments.



**Figure 1:** Total number of rabbit shipments by month for 2007



**Figure 2:** Number of rabbit behavior complaints by month for 2007

Complaints received about the behavior of the Dutch belted rabbits focused on their stress to new environments and aggression towards human caretakers. Since rabbit aggression against humans stems primarily from fear, the socialization program focused on introducing the rabbits to common but novel stimuli early in their life to decrease an overall fear response (Crowell-Davis, 2006). The socialization program employed with this colony exposed rabbit pups and weanlings to animal handling techniques commonly employed in research facilities in the United States such as physical exams, toe nail trims, placement in physical restrainers, placement on a smooth surface for physical manipulation and objects commonly encountered such as latex gloves and scales (Figure 3). Additionally, commonly used environmental enrichment devices such as rattlers, balls, dumbbells, and fruit treats (Bio-Serv, 2007, Harris *et al.*, 2001; Johnson *et al.*, 2003) were introduced to the rabbits at a relatively early age so that these devices early so that as adults the objects will not elicit as much of a fear response (Pongrácz *et al.*, 2001). The caretakers in this pilot study noted that the use of the fruit treats increased the animals' "non-freezing" or "bold" behaviors as defined by Jezierski and Konecka (1996).



**Figure 3:** Object introduction

## CONCLUSIONS

Dutch belted rabbits are commonly used as research animals due to their small size compared to New Zealand white rabbits. However, in the United States, it has been reported that commercially raised black Dutch belted rabbits are more aggressive towards their human handlers and less adaptive to environmental change than their New Zealand White counterparts. Specific complaints were generated regarding the behavior of black Dutch belted rabbits from one commercial colony. This pilot study was novel because it focuses exclusively on the black Dutch belted rabbit as a research model when the vast majority of behavior data is focused on laboratory New Zealand whites.

An intensive socialization program was initiated in a colony of black Dutch belted rabbits raised exclusively for research after the receipt of a cluster of complaints regarding behavior. This program includes adaptation to common animal handling procedures, environmental enrichment devices, and other materials encountered in the laboratory. This program encompasses handling pre- and post-weaning and prepares the animals for use in research with less overall aggression to human caretakers and less stress to novel environmental stimuli. Preliminary success of the program has been demonstrated due to a decrease in the number of complaints received about animal behavior once the rabbits have arrived at the research facility.

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