



## Canine Research

## Horn of calf hooves as chews in laboratory dogs



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## ABSTRACT

European law recommends the enrichment of laboratory dogs with chewing objects. This study examines the suitability of the horn of calf hooves for this purpose and was performed with 62 dogs (60 beagles, 2 Foxhound Boehringer Ingelheim) across 3 research facilities. In facility A, 18 female dogs and 1 neutered male were housed in 6 groups, in facility B, 16 intact male dogs were housed in 8 groups and in facility C, 25 female dogs were housed in 6 groups under their usual housing conditions. Each group of dogs received autoclaved calf horn for 3 consecutive days. In addition to its role as an enrichment object, the nutritional components of the horn were determined. Dogs in facilities A and C chewed intensively in the first hour after they received the horn (average for A = 29.3 minutes, average for C = 22.8 minutes per dog). Thereafter, the chewing time decreased during the day. In facility B, many dogs initially reacted by barking and jumping at the horn, suggesting a reaction of uncertainty, most likely due to the dogs' young age and lack of experience. Dogs in facilities A and C chewed significantly longer ( $P = 0.009$  in A;  $P = 0.000$  in C) and more frequently ( $P = 0.008$  in A,  $P = 0.001$  in C) on the first day compared to the second day. Consistent with these findings, the daily weight loss of the horn in facilities A and C was significantly higher on the first day compared to the second day ( $P = 0.003$  in A,  $P = 0.002$  in C). Fights over the horn were observed in 2 groups of intact male dogs, aged 4–6 months in facility B. Injuries of the oral cavity were not observed. Two dogs of facility B had minor crown fractures of a deciduous *dens caninus*. In conclusion, autoclaved horn of calf hooves is an attractive chewing object for dogs after a change in deciduous teeth. However, dogs should be supervised when these enrichments are newly introduced.

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## Introduction

Enrichment in laboratory animal husbandry is recommended by the European Directive 2010/63/EU (2010). Accordingly, enrichment should extend the range of activities, including physical exercise, foraging, and manipulative behaviors. Special recommendations for laboratory dogs are given in Appendix A of ETS No. 123 (Council of Europe, 2006). One of the demands listed is “as chewing is an important behavior, items should be provided which meet this need.”

The Joint Working Group on Refinement (2004) also emphasizes the importance of chewing for dogs. Baumans and Van Loo (2013) stressed the importance of evaluating enrichment objects for laboratory animals. However, for laboratory dogs, there is still no standard chewing object that has been approved for both acceptance and suitability. Hubrecht (1993, 1995) found that laboratory puppies and young dogs readily engaged with toys and chewing objects that were attached to chains hanging from the ceiling. James (1961) described that puppies preferred soft over hard objects. These findings were extended by Pullen et al. (2010), showing that kenneled dogs rarely used robust toys, but engaged with toys, such as teddy bears and tennis balls, which they could crunch. Moreover, the dogs preferred toys lying on the floor rather than toys that were hanging freely. In addition, DeLuca and Kranda (1992) described that laboratory dogs chewed up their toys. However, toys that can be chewed up bear considerable health risks.

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Particles and sharp objects can result in injuries of the mouth and/or the digestive tract. Although DeLuca and Kranda (1992) did not report gastrointestinal problems, this risk exists.

In comparison to companion dogs, laboratory dogs are not under permanent supervision. Thus, toys and chewing objects used in a research facility must meet high safety standards. The Joint Working Group on Refinement (2004) recommends that chewing objects should be carefully selected “to avoid the (rare) risk of mouth ulceration or blocking of the gut.” To avoid the danger of swallowing plastic, wood, or rubber, digestible chewing objects should be used. Previous studies of Hubrecht (1995; 1993) showed that puppies and young dogs clearly preferred rawhide to plastic or wooden toys. A research facility which participated in our study successfully used the horn of calf hooves, hereafter named calf horn, for several years. On the basis of these experiences, the present study aimed to examine the suitability of calf horn as enrichment objects for laboratory dogs as well as its nutritional components.

## Materials and methods

### Pretest

To obtain an overview of calf horn usage, three 5-year-old intact male beagles received the horn of 4 calf hooves for a period of 77 hours. Video recordings were evaluated with behavior sampling and continuous recording (Martin and Bateson, 2007) during the entire period. All feces were collected every morning and suspended in water. Horn fragments were extracted by sieving. At the end of the observation period, the mouths of the dogs were examined.

### Animals and husbandry

The main experiment was performed on a total of 62 purpose-bred laboratory dogs (60 Beagles, 2 Foxhound Boehringer Ingelheim) in 3 facilities (A, B, and C). The dogs remained in their usual, socially stable groups in the environment that they were accustomed to and were fed once a day with dry complete and balanced diet (Josera, Kleinheubach, Germany) soaked in drinking water in facility A, with dry food (“PROVIMI KLIBA SA,” Gosau, Switzerland) in B, and with freshly prepared food according to the facility’s own recipe in C.

In A, 18 female and 1 neutered male beagle and 2 female Foxhound Boehringer Ingelheim dogs, 1.5 to 10.6 years old, (average 4.5 years) were kept in 6 groups of 2 to 4 dogs. During the daytime, the dogs were kept in enriched open-air runs (29 m<sup>2</sup> and 54 m<sup>2</sup>). At night, they were housed in groups in indoor kennels (6 m<sup>2</sup>) equipped with a plastic bed for each dog. In the runs, the dogs were provided with wooden branches, cotton ropes, tennis balls, and biweekly with the horn of calf hooves.

In B, sixteen 4- to 10-month-old intact male beagles (average 0.6 years) were kept in eight groups of 2 beagles, in indoor kennels (11.2 m<sup>2</sup>), equipped with an elevated plastic board as resting place. The caretakers played with every group twice a week using dog toys. Chewing objects had not been used as enrichment. In C, 25 intact 9-month to 6.3-year-old female beagles (average 2.0 years) were kept in 5 groups of 4 dogs and 1 group of 5 dogs in indoor kennels (of 7.0 m<sup>2</sup>) equipped with rubber mats. The dogs had permanent access to outdoor runs (18.5 m<sup>2</sup>). The caretakers played with the dogs once a week with dog toys. The dogs received cow bones once a week.

### Presentation of calf horn

The calf horn shown in Figure 1 (Barabas GmbH, Kerken, Germany) was autoclaved in a steam sterilizer (Tecnomara



Figure 1. Horn from the calf hoof.

Deutschland GmbH) for 30 minutes at 121 °C and given to the dogs on 3 consecutive days (Tuesday to Thursday) from 8 to 15 hours in A, from 8 to 16 hours in B and from 7:30 to 15:00 hours in C.

On day 1, groups of 2 dogs received 3 horns, groups of 3 dogs received 5 horns, groups of 4 dogs received 6 horns, and groups of 5 dogs received 8 horns. On the following 2 days, the remaining pieces weighing more than 6 g were given. Facility C requested new horns on day 3 to ensure that there were always more horns than dogs per group.

### Video recordings

The groups were recorded during the daytime while the horn was available with cameras covering the dogs’ kennels and runs. The 5× time-lapse recordings from day 1 and day 2 were evaluated with behavior sampling and continuous recording (Martin and Bateson, 2007). Every use of the calf horn of at least 30 seconds as defined in Table 1 was noted with its starting and end points. Chewing, barking, or jumping at the horn for less than 30 seconds was recorded as frequency of use. The videos of day 3 were only screened regarding possible scuffles.

### Soiling and usage of the calf horn

Every afternoon, at 15 hours in A and C, and at 16 hours in B, all of the calf horn objects were collected, weighed, and the degree of soiling was evaluated using a scoring system of 0 = no visible

Table 1  
Behavioral types of calf horn use

Behavior	Definition
Chewing	Chewing on the horn with the teeth
Barking/jumping at	Barking while directly looking at the horn and/or jumping at the horn: jumping motion in the direction of the horn with lifting on or more paws
Burying	Burying motion with paws in close proximity of the horn
Holding with paws	Holding the horn with the paws without chewing on it (≥30 seconds)
Under the board with horn (only in B)	Dog is under the lying board with the horn and cannot be seen (≥30 seconds)

Definition of behavioral types of calf horn usage; video analysis via behavior sampling and continuous recording (Martin and Bateson, 2007).

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