World Rabbit Sci. 2021, 29: 109-114
doi:10.4995/wrs.2021.14193
WRSA, UPV, 2003

# CARDBOARD AND RUBBER OBJECTS AS MEANS OF ENVIRONMENTAL ENRICHMENT FOR RABBITS

DA SILVA K.G.*, STANGE M.G*, PERGORARA M.†, SOTOMAIOR C.S.*, WEBER S.H.*, BORGES T.D.*, COSTA L.B.*

*Graduate Programme in Animal Science, Pontifícia Universidade Católica do Paraná (PUCPR), Imaculada Conceição, 1155, Curitiba, Paraná, 80215-901, Brazil.
†Undergraduation in Veterinary Medicine, Pontifícia Universidade Católica do Paraná (PUCPR), Imaculada Conceição, 1155, Curitiba, Paraná, 80215-901, Brazil.

Abstract: Environmental enrichment improves rabbit welfare in rabbitries. Various toys for cats and dogs are commercially available, which are made of materials that could be safely used for rabbits as well. The objectives of this study were to evaluate whether cardboard and rubber materials could be used for environmental enrichment for rabbits. The study involved 42 adult New Zealand white rabbits (20 females and 22 males), randomly assigned to seven treatment groups: "C", without object (control); "RB", a solid rubber ball; "FT", a fillable teether filled with hay; "CH", a cardboard hole; "CS", a piece of a cat scratcher; "CSC", a piece of a cat scratcher with catnip; "CF", an articulated cardboard fish. The behaviour of the rabbits and the percentage of destruction of the objects were recorded for 28 d. The normal behaviours of locomotion, rearing, stretching, stereotypies and sitting were not influenced by the treatments. Lying down was observed more frequently than the full stretched out position for resting. The FT-treatment group presented most behaviours of interaction (biting and sniffing) ($P<0.05$) as compared to RB, CSC, and CF-treatment groups. All the objects showed some level of destruction; the mean rates of destruction for CH, CS, CSC and CF were up to 40%, whereas those for FT and RB were under 30%. Taken together, the results suggest that cardboard and rubber materials can be used as means of environmental enrichment for rabbits.

Key Words: rabbits, behaviour, *Oryctolagus cuniculus*, welfare.

## INTRODUCTION

Environmental enrichment techniques related to the expression of species-typical behaviours such as chewing, rearing and exploring may improve rabbit welfare (Bayne, 2003). In rabbit production, the most common applications of environmental enrichment are for does and growing rabbits (Rommers *et al.*, 2014; Bozicovich *et al.*, 2016). In general, there are few studies on environmental enrichment for adult males and females (Hansen and Berthelsen, 2000), except in laboratory animals (Lidfors, 1997; Poggiagliolmi *et al.*, 2011). However, environmental enrichment for these animals is recommended, as they are also prone to boredom and stereotypies (Gunn and Morton, 1995).

Chewing objects are desirable as rabbit environmental enrichment objects, as chewing is a species-typical behaviour (Bayne, 2003). Different materials have been used as enrichment for growing rabbits, such as wood sticks (Princz *et al.*, 2007), iron chains (Lang *et al.* 2011), rubber, cardboard and food for laboratory rabbits (Harris *et al.*, 2001; Poggiagliolmi *et al.*, 2011). Some of these objects are not commercially available and are difficult to acquire.

Correspondence: L.B.Costa, *batista.leandro@pucpr.br.* Received *August 2020* - Accepted *January 2021.*
https://doi.org/10.4995/wrs.2021.14193

Various products for cats and dogs are made of cardboard and rubber, which would be an option for use with rabbits. The objectives of this study were to evaluate whether cardboard and rubber objects improve welfare of rabbits on normal behaviour, activity/inactivity, abnormal behaviour, object attractiveness and the object's destruction.

## MATERIALS AND METHODS

### *Animals and husbandry conditions*

The study was conducted in the rabbitry of the University Experimental Farm of Pontifícia Universidade Católica do Paraná (PUCPR) in Fazenda Rio Grande, Paraná, Brazil, between March and April of 2019. It was approved by the Animal Use Committee of PUCPR, under protocol number 01267.

The study involved 42 New Zealand white rabbits (20 females and 22 males) more than 6 mo old. The body weight average was 4.667±0.627 kg and ranged from 3.4 to 6.1 kg. They received commercial feed, water and hay *ad libitum,* and were housed individually in suspended wire cages [80 cm(L)×60 cm(W)×40 cm(H)] with automatic water dispensers, manual feeders and hay racks. The lighting pattern was natural, with average environmental temperature of 20±3.6°C (maximum: 30°C, minimum 13°C), humidity average of 70±14.6% (maximum: 100%, minimum 29%) and a photoperiod of 12h:12h. All rabbits, males and females were not mated during the experiment.

### *Experimental protocol*

The rabbits were randomly assigned to seven treatment groups (six animals per group and the animal is the experimental unit, n=6): "C", no new object in the cage (control); "RB", a solid rubber ball with artificial scent of banana (diameter 5 cm; Macaquinho, Pet Games®); "FT", a fillable teether (maximum/minimum diameter of 7 cm/5 cm×10 cm (H); Monstrinho, Pet Games®) filled with the same hay as that used for feeding; "CH", a cardboard hole [40 cm (L)×25 cm (H)×21 cm (W); Toca do gato, Pet Games®]; "CS", a 6cm piece (L)×5 cm (H)×6 cm (W) of cat scratching post (Cat Tower, Pet Games®); "CSC", a 6 cm piece (L)×5 cm (H)×6 cm (W) of cat scratching post (Cat Tower, Pet Games®), sprayed, on day-0 (D0), with 4 spray pumps of catnip (Catnip Spray, Pet Clean®) on each side; "CF", an articulated cardboard fish with elastic cord (Eco caça peixe, 13 cm (L)×6 cm (H)×5 cm (W), 39 g, Pet Games®). Each rabbit participated in only one treatment for the entire experimental period. The CH, CSC/CS, and CF objects are toys recommended for cats; RB and FT objects are toys recommended for dogs. All objects are commercially available in Brazil (Figure 1).

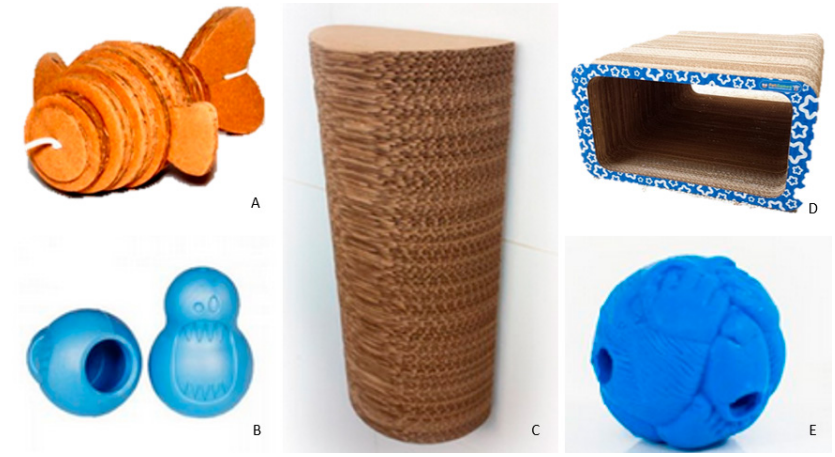


Figure 1: Objects used in the experiment. A= articulated cardboard fish with elastic cord, Pet Games® (CF group); B= fillable teether, Pet Games® (FT group); C= cat scratching post, Pet Games® (CS and CSC groups); D= cardboard hole, Pet Games® (CH group); E= a solid rubber ball with artificial scent of banana, Pet Games® (RB group).

The CF was fixed by its cord to the central part of the cage roofs, at a distance of 20 cm from the floor. If the rabbit pulled out the object and destroyed it, or it dropped from the cage, the object was not replaced, and the evaluation was concluded. The RB, CH, CS, and CSC treatments had their objects placed inside the cage, without interference from evaluators during all the experiments (Figure 2). The FT object was filled with hay every day, 5 min before the evaluations.

### *Ethogram, behavioural findings and object destruction*

Before starting the experiment, training to recognise rabbit behaviour in the rabbitry was undertaken regarding the studies of Gunn and Morton (1995), Princz *et al.* (2007), Poggiagliolmi *et al.* (2011) and Rommers *et al.* (2014). Rabbit behaviours were classified as self-cleaning, yawning, stretching, eating, drinking, sitting, lying down (sternal and stretched), standing, in movement, biting the cage, biting the nipple, digging the grid, walking in circles, caecotrophy and object-related behaviours: sniffing, gnawing, pulling, eating, licking, and marking via the chin scent glands. Subsequently, a standard list of behaviours to be analysed during the experiment was defined (Table 1).

All the objects were placed on D0, with observations commencing on day 1 (D1) until day 28 (D28). The behavioural evaluation was conducted daily for 28 d, from 14:00 to 16:30 h, by one observer, with two trained observers alternately taking each observation day. The evaluator stood in front of two cages for 3 min (adapted from Rommers *et al.*, 2014) with behaviours being recorded as presence/absence (Table 1). At the end of the experiment, all presences of a behaviour were accounted; the same was done for absence of each behaviour. Then, the average presence of each behaviour was obtained.

The destruction of objects was analysed using the percentage of destruction and number of days into the experiment. The percentage of destruction of the objects, from 0 to 100%, was visually evaluated until day 27. For objects destroyed to 100%, behaviours were evaluated up until the objects were completely destroyed, with no further evaluations of behaviour for that object for the remainder of the experiment.

To analyse the activity/inactivity of the rabbits, the behaviours of lying down, sitting, rearing, fully stretched out, and locomotion were included in analysis (Table 1). The evaluation of abnormal behaviours was performed using data on stereotypies (Table 1). Fully stretched out and stretching behaviours were interpreted as indicators of relaxation (Buseth and Saunders, 2018). Other normal behaviours, such as eating feed and hay, drinking, grooming and caecotrophy, were included as normal behaviours (Rommers *et al.*, 2014) (Table 1).

### *Statistical analysis*

The behaviour data was submitted to the Kruskal-Wallis test. For evaluations of rabbits lying down, being fully stretched out, biting the objects and sniffing the objects, the Mann-Whitney test was applied to analyse significant

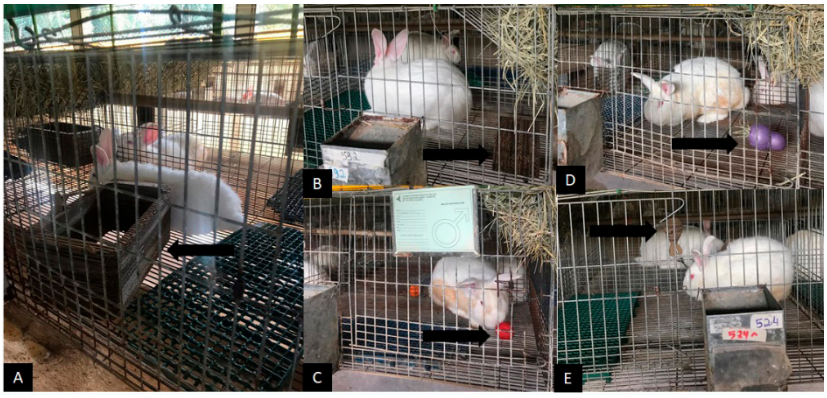


Figure 2: Positioning of the objects in the cage. A=CH group; B=CS and CH groups; C=RB group; D=FT group; E=CF group.

Table 1: Behavioural patterns of rabbits observed during 3-min evaluation.

| Type | Behaviour | Definition |
|---|---|---|
| Normal | | |
| | Eating feed | Consuming commercial feed |
| | Eating hay | Consuming hay |
| | Drinking | Consuming water |
| | Caecotrophy | Consuming caecotrophes |
| | Grooming | Licking or scratching the coat |
| Activity/Inactivity | | |
| | Lying down | Lying down in sternal recumbent |
| | Full stretched out | Resting with ventral contact of the torso, with hind legs stretched out. |
| | Sitting | Supported with the four members on the floor of the cage, with no contact of the torso |
| | Rearing | Animal upright, with hind legs in contact with the floor and forelimbs not supported |
| | Locomotion | Movement around the cage (walking, jumping, running) |
| | Stretching | Extending limbs to their full extend |
| Abnormal | | |
| | Stereotypy – biting cage | Repetitive behaviour of biting any cage structure (wire, feeder, drinker) for, at least, 90 s |
| | Stereotypy – digging | Repetitive behaviour of digging on the wire of cage for, at least, 90 seconds |
| | Stereotypy – circling | Repetitive behaviour of circling movement around the cage for, at least, 90 seconds |
| Attractiveness * | | |
| | Sniffing | Sniffing or chin-mark the object |
| | Biting | Gnawing, biting or eating the object |

*not observed in the control group (C)

differences. The destruction rates were analysed using analysis of variance (ANOVA). Statistical analysis was carried out using IBM SPSS Statistics 24, with a 5% significant level ($P<0.05$).

## RESULTS AND DISCUSSION

The normal behaviours did not significantly differ ($P>0.05$) among different treatment groups (Table 2). The time of evaluation (14:00 to 16:30 h) could explain the minor feeding behaviour observations (eating feed, eating hay, and drinking) and the higher percentages of grooming as resting-related behaviours (less locomotion and more lying down). The normal behaviours of rabbits are generally not affected by environmental enrichment (Poggiagliolmi *et al.*, 2011; Rommers *et al.*, 2014), although a variation was observed by Princz *et al.* (2007) and Bozicovich *et al.* (2016).

The treatments did not influence locomotion, rearing and sitting behaviours. The objects that contained or resembled feed (FT, RB, and CSC) had no significant effect on activity/inactivity; however, Harris *et al.* (2001) observed a relationship between feed enrichment and behavioural activity of rabbits. A possible explanation for the non-significant change in behaviour of rabbit that received a food-related object was that all animals received hay during the experimental period. According to Berthelsen and Hansen (1999), hay could be considered an enrichment for rabbits. The period of observation (14:00 to 16:30 h) of the animals could also have contributed to these results, as the observations were done in the afternoon, a period which the animals are less active.

Fewer lying down behaviours were observed in the C, CS and RB treatment groups, where 32.7 and 29.8% of rabbits in RB and CS treatment groups, respectively, presented the fully stretched out position. This position was described as a resting position in relaxed rabbits (Buseth and Saunders, 2018). No significant difference was found in stretching behaviour among all the treatment groups.

The treatments did not have a significant effect on stereotypies. The time of evaluation and hay feeding could explain the lower presence of stereotypies of rabbits, as described above.

The rabbits that received FT presented the most interactive behaviour with objects (biting and sniffing), as the hay inside the toy is attractive to rabbits (Lidfors, 1997). The FT object attracted the attention of the rabbits even with the

Table 2: Mean percentage of rabbits' behaviour, according to treatments

| | | Treatments | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Evaluation | Behaviour (%) | CH | FT | RB | CSC | CS | CF | C | *P*-value |
| Normal | | | | | | | | | |
| | Eating feed | 4.8 | 3.6 | 6.5 | 6.7 | 7.7 | 6.5 | 7.7 | 0.667 |
| | Eating hay | 0.6 | 4.2 | 4.2 | 3.6 | 1.8 | 1.6 | 3.6 | 0.298 |
| | Caecotrophy | 0.6 | 3.0 | 1.8 | 2.4 | 0.6 | 0.8 | 3.0 | 0.368 |
| | Drinking | 6.0 | 0.6 | 1.8 | 3.0 | 2.4 | 0.8 | 3.0 | 0.052 |
| | Grooming | 30.4 | 32.1 | 22.6 | 27.3 | 28.6 | 31.7 | 31.0 | 0.509 |
| Activity/Inactivity | | | | | | | | | |
| | Locomotion | 23.8 | 20.2 | 20.2 | 16.4 | 18.5 | 15.4 | 15.5 | 0.423 |
| | Lying down | $67.9^{bc}$ | $74.4^{c}$ | $56.5^{a}$ | $70.3^{bc}$ | $61.3^{ab}$ | $70.7^{bc}$ | $64.3^{ab}$ | 0.010 |
| | Full stretched out | $21.4^{ab}$ | $17.9^{a}$ | $32.7^{c}$ | $21.2^{ab}$ | $29.8^{bc}$ | $17.1^{a}$ | $25.6^{abc}$ | 0.005 |
| | Sitting | 37.5 | 48.2 | 34.5 | 41.8 | 39.9 | 44.7 | 44.0 | 0.188 |
| | Rearing | 9.5 | 8.3 | 6.0 | 7.3 | 8.3 | 6.5 | 11.3 | 0.634 |
| | Stretching | 4.2 | 4.8 | 6.0 | 7.9 | 5.4 | 8.9 | 4.8 | 0.540 |
| Abnormal | | | | | | | | | |
| | Stereotypy biting cage | 1.2 | 3.0 | 1.8 | 0.6 | 1.2 | 4.9 | 1.8 | 0.171 |
| | Stereotypy digging | 1.2 | 1.8 | 0 | 1.2 | 0.6 | 0.8 | 1.2 | 0.766 |
| | Stereotypy circling | 0 | 0 | 0 | 0.6 | 0 | 0 | 0 | 0.442 |
| Attractiveness | | | | | | | | | |
| | Sniffing | $6.5^{ab}$ | $9.5^{b}$ | $3.6^{a}$ | $2.4^{a}$ | $5.4^{ab}$ | $2.4^{a}$ | - | 0.001 |
| | Biting | $6.0^{b}$ | $18.5^{c}$ | $0.6^{a}$ | $1.8^{ab}$ | $3.0^{bc}$ | $0^{a}$ | - | <0.001 |

[a-c] Different letters, in the same line, are different between them at $P<0.05$.

CH: cardboard hole; FT: fillable teether filled hay; RB: rubber ball with artificial scent of banana; CSC: piece of a cat scratcher with catnip; CS: piece of a cat scratcher; CF: articulated cardboard fish; C: control.

provision of hay as food on the hay rack. The scent of banana (RB) and catnip (CSC) was not sufficient to induce more interaction behaviours during the observations; likewise, they were not repellent for the animals, as their destruction was observed. In a study by Poggiagliolmi *et al.* (2011), preference between cardboard and rubber materials was not observed, even with fruit flavoured cardboard material.

All cardboard objects (CH, CS, CSC, CF) were destroyed with a mean destruction rate of up to 40% (Table 3). The destruction rate of CF was 54%, with 3 objects (50%) being completely destroyed. Furthermore, the durability of the objects was 6 d. For CSC and CS objects, 66.7% of them had low destruction, with a durability of 22 and 26 d, respectively. The CH objects, 66.7%, showed medium to high destruction, with a durability of 23 d. Suspended objects, such as CF, could induce the natural behaviour of rearing (Bayne, 2003); this behaviour could have contributed to the

Table 3: Destruction (%) of objects by rabbits, according to treatments.

| Treatment | Destruction* (mean±SD) % | No destruction % | Low destruction % | Medium/High destruction % | Total destruction % |
|---|---|---|---|---|---|
| RB | 28±38.1 | 50.0 | 0 | 50.0 | 0 |
| FT | 0±0.8 | 83.3 | 16.7 | 0 | 0 |
| CH | 40±30.1 | 0 | 16.7 | 66.7 | 16.7 |
| CS | 40±36.3 | 0 | 66.7 | 16.7 | 16.7 |
| CSC | 41±46.1 | 0 | 66.7 | 0 | 33.3 |
| CF | 54±50.2 | 0 | 50.0 | 0 | 50.0 |

There were no significant difference between averages of destruction (*$P>0.05$). No destruction: object no destructed; Low destruction: object with <25% of destruction; Medium/high destruction: object with 25 to 99% of destruction; Total destruction: object with 100% of destruction.

RB: rubber ball with artificial scent of banana; FT: fillable teether filled hay; CH: cardboard hole; CS: piece of a cat scratcher; CSC: piece of a cat scratcher with catnip; CF: articulated cardboard fish. SD: standard deviation.

higher destruction rate and low durability of the CF object and the probability that this interaction occurred outside of observations.

More than 50% of rubber objects remained intact at the end of the experiment; with mean values of destruction below 30%. The thermoplastic constituent of rubber objects was resistant to rabbit interaction (Poggiagliolmi *et al.*, 2011); moreover, FT objects were better preserved because the hay inside them was the focus of the animals. The rabbits that received the banana flavoured object (RB) could have had more interaction with the object, although this was not observed during the evaluations.

The feeding system appears to have influenced the rabbits' responses to the inclusion of objects. The interaction with the objects decreased when rabbits received an amount of alfalfa hay (Lang *et al.*, 2011). Hay is a known effective environmental enrichment for rabbits (Lidfors, 1997). However, even in the presence of hay, interactions with objects were observed, such as biting, sniffing or destroying the enrichment objects. The FT object with hay appeared to be more attractive to the rabbits. The use of catnip did not induce interaction with the cardboard, as observed interaction and destruction rates were not significantly different between CSC and CS. All cages with cardboard materials (CSC, CS, CF, and CH) had some destruction, with rubber objects (RB, FT) appearing to be more resistant. In conclusion, cardboard and rubber materials can be used as environmental enrichment for rabbits.

Acknowledgements: This study was financed in part by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brasil (CAPES) - Finance Code 001. We thank the PUCPR for the scholarship and Petgames® for donation of the toys.

## REFERENCES

Bayne K.A. 2003. Environmental enrichment of nonhuman primates, dogs and rabbits used in toxicology studies. *Toxicol. Pathol., 31: 132-137. https://doi.org/10.1080/01926230390175020*

Berthelsen H., Hansen L.T. 1999. The effect of hay on the behavior of cages rabbits (*Oryctolagus cuniculus*). *Anim. Welfare, 8: 149-157.*

Bozicovich T.F.M., Moura A.S.A.M.T., Fernandes S., Oliveira A.A., Siqueira E.R.S. 2016. Effect of environmental enrichment and composition of the social group on the behavior, welfare, and relative brain weight of growing rabbits. *Appl. Anim. Behav. Sci., 182: 72-79. https://doi.org/10.1016/j.applanim.2016.05.025*

Buseth M.E., Saunders R.A. 2018. Rabbit behavior, health and care. *CABI, Boston, USA.*

Gunn D., Morton D.B. 1995. Inventory of New Zealand White rabbits in laboratory cages. *Appl. Anim. Behav. Sci., 45: 277-292. https://doi.org/10.1016/0168-1591(95)00627-5*

Hansen L.T., Berthelsen H. 2000. The effect of environmental enrichment on the behaviour of caged rabbits *(Oryctolagus cuniculus). Appl. Anim. Behav. Sci., 68: 163-178. https://doi.org/10.1016/s0168-1591(00)00093-9*

Harris L.D., Custer L.B., Soranaka E.T., Burge J.R., Ruble G.R. 2001. Evaluation of objects and food for environmental enrichment of NZW rabbits. *J. Am. Assoc. Lab. Anim. Sci., 40: 27-30.*

Lang C., Weirich C., Hoy S. 2011. Frequency of occupation with different objects by growing rabbits under various conditions. *J. Agr. Sci. Tech- Iran., 10: 1-9.*

Lidfors L. 1997. Behavioral effects of environmental enrichment for individually caged rabbits. *Appl. Anim. Behav. Sci., 52: 157-169. https://doi.org/10.1016/S0168-1591(96)01141-0*

Poggiagliolmi S., Crowell-Davis S.L., Alworth L.C., Harvey S.B. 2011. Environmental enrichment of New Zealand White rabbits living in laboratory cages. *J. Vet. Behav., 6: 343-350. https://doi.org/10.1016/j.jveb.2010.12.001*

Princz Z., Orova Z., Nagy I., Jordan D., Stuhec I., Luzi F., Verga M., Szendrő Zs. 2007. Application of gnawing stick in rabbit housing. *World Rabbit Sci., 15: 29-36. https://doi.org/10.4995/wrs.2007.607*

Rommers J.M., Bracke M.B.M., Reuvekamp B., Gunnink H., De Jong I.C. 2014. Cage enrichment: rabbit does prefer straw or a compressed wooden block. *World Rabbit Sci. 22: 301-309. https://doi.org/10.4995/wrs.2014.1353*