

**Use of collard green stalks as environmental enrichment for cockatiels
(*Nymphicus hollandicus*) kept in captivity**

[Utilização de talos de couve como enriquecimento ambiental para calopsitas
(*Nymphicus hollandicus*) mantidas em cativeiro]

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ABSTRACT

The objective of this study was to evaluate the influence of collard green stalks as environmental enrichment on the behavior of cockatiels (*Nymphicus hollandicus*) maintained in captivity. Sixteen birds were housed individually in cages and fed a commercial diet for parrots and seed mix, with water ad libitum during 12 days in climatic chamber with temperature and relative humidity simulating tropical conditions. The animals (n = 8) were divided into two groups, control and enriched, and the experimental period was divided into two phases (during and after enrichment) of four days each. The collard green stalks were used as enrichment only during the first period of the experiment, daily changed and maintained only during the day. The stalks increased the feed intake and decreased the sleep activities. No effect was observed on the body surface temperature, locomotion, maintenance and other resting activities. Small undesirable activities were detected. It was concluded that collard green stalks can be used as environmental enrichment, becoming part of food preferences of cockatiels. However, its use does not significantly alter the behavior of birds kept in captivity

Keywords: birds, animal welfare, captive animals, food enrichment, psittacine

RESUMO

O objetivo deste estudo foi avaliar a influência de talos de couve como enriquecimento ambiental sobre o comportamento de calopsitas (*Nymphicus hollandicus*) mantidas em cativeiro. Dezesesseis aves foram alojadas individualmente em gaiolas e alimentadas com dieta comercial para psitacídeos e mistura de sementes, com água ad libitum durante 12 dias, em câmara climática com temperatura e umidade relativa do ar simulando condições tropicais. Os animais (n=8) foram divididos em dois grupos, controle e enriquecido, e o período experimental foi dividido em duas fases (durante e após o enriquecimento) de quatro dias cada. Os talos de couve foram usados como enriquecimento apenas durante o primeiro período do experimento, trocados diariamente e mantidos apenas durante o dia. Os talos aumentaram o consumo de ração e diminuíram o comportamento dormir. Nenhum efeito foi observado nas outras atividades de descanso, na temperatura da superfície corporal, na locomoção e manutenção. Foram detectadas pequenas atividades indesejáveis. Concluiu-se que os talos de couve podem ser usados como enriquecimento ambiental, formando parte das preferências alimentares de calopsitas. No entanto, seu uso não altera de maneira expressiva o comportamento das aves.

Palavras-chave: pássaro, bem-estar animal, animais cativos, enriquecimento alimentar, psitacídeo

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INTRODUCTION

Recently, the use of cockatiels (*Nymphicus hollandicus*) as pets is increasing around the world. However, studies with environmental enrichment and particularly on its influence on the behavior of this species are scarce. Due to an increase in the number of animals maintained in captivity, there is a great interest in the behavior of these birds when maintained with different substrates as environmental enrichment (Meehan and Mench, 2002; Meehan *et al.*, 2004). This knowledge contributes with future studies in order to improve captivity conditions of the animals.

Environmental enrichment can be defined as modification of the environment of captive animals, in order to increase opportunities for expression of biological behavior and improve the health conditions of animals (Newberry, 1995). However, to provide adequate environment, it is necessary to know the behavior of species maintained in different conditions. Cockatiels are mostly characterized as docile animals but different behavioral manifestation can be expressed when the birds are submitted to different environmental conditions (Seibert and Crowell-Davis, 2001).

There is evidence that birds can interact with several types of plants in the environment (Del Hoyo *et al.*, 1997). In the literature, no study has reported on the use of collard green stalks (*Brassica oleracea L.*) as environmental enrichment on the behavior of captivity birds. In general, collards are considered to be a healthy food for humans (Chen *et al.*, 2014), being a good source of vitamins (A, C, K and folic acid), calcium and soluble fiber, among several others nutrients like iron, copper, manganese, selenium and zinc (Miller-Cebert *et al.*, 2009; Chen *et al.*, 2014).

The knowledge of the effects of the use of collard green stalks as environmental enrichment for captivity cockatiels would assist in the discovery of new information that would improve the life quality of the birds. Thus, the aim of this study was to evaluate the use of collard green stalks as environmental enrichment for cockatiels (*N. hollandicus*) kept in captivity.

MATERIAL AND METHODS

The research was conducted at the Department of Animal Science of Federal University of Lavras (UFLA), in Lavras, Minas Gerais, Brazil. The experimental protocol was approved by the Ethics Committee on Animal Use (CEUA) of the same institution (protocol number 029/13).

Sixteen healthy cockatiels (*N. hollandicus*), eight males and females, aged approximately one year, derived from commercial breeding, were housed individually in galvanized wire cages (38×37×28cm), separated by 20cm from each other, at 1.0m of the floor, in a climatic chamber equipped with an automated system (MT-530 Super, Full Gauge Controls, Canoas, Brazil) that controlled the circulation of heated or cooled air in the room using infrared lamps, blowers and fans. All the equipment in a climatic chamber was connected to a central panel, allowing the automatic adjustment of the internal temperature of the room. The experimental conditions were adjusted to maintain the temperature near to 35°C from 6h00min to 18h00min and 25°C from 18h01min to 5h59min. The relative humidity was set to 70%. This change in day and night temperatures was determined based on the natural temperature variations that occur in the tropical zone (Verstappen *et al.*, 2002).

The air in the chamber was changed constantly and was regulated by blowers and fans attached to a pipe with small holes for air distribution. The system automatically stayed on for 15 minutes and off for two minutes. The temperature was monitored daily inside the rooms at 8h00min, 13h00min and 18h00min using maximum and minimum thermometers placed at half the height of the animals. The lighting program used was 24 hours of artificial light with infrared light 250w (Ecolume, Belo Horizonte, Brazil) located at 1.0m above the birds.

Each cage contained two plastic feeders, one for commercial diet (Complete Food for Parrots Birds CC Bio Tron, Rio Claro, São Paulo, Brazil) and other for seed mix (50% millet, 30% canary seed, 15% oat and 5% sunflower - Torloni, 1991), and a drinking ceramic pot (10 cm diameter). Excreta were daily collected in an aluminum tray located under each cage.

The experiment was carried out in a completely randomized design with eight replicates in split-plot in time scheme (during and after the use of collard green stalks) with two treatments (with or without collard green stalks). The experimental unit was one bird evaluated during four days.

Initially, the animals were divided into two groups of eight birds each (four males and four females) that received or not the collard green stalks as environmental enrichment. The groups were organized so as to be the most homogeneous possible (size, weight and previous behavior such as aggression). The experimental period (8 days) was divided into two periods of four days (during and after the enrichment). The birds received water and diets *ad libitum*. Handlings (cage cleaning, feeding, water change, and evaluation of consumption) were made daily always around 6h00min. The feed and seed intake was determined based on the difference between the amount provided and the waste.

The environmental enrichment was made with collard green stalks during the first period of experiment (from 1st to 4th day). The stalks were heavy and then put in each cage of enriched group twice, at 6h00min and 13h00min. At 18h00min, the leftover stalks were removed and weighed again to evaluate consumption of the material. The body temperature was daily measured using the device FLIR E50 (Nashua, USA) around at 6h00min, 12h00 and at 18h:00min, and the daily mean calculated.

The behavior of the birds was assessed by filming during daylight with a camcorder (8.0 mm CCD Color Camera 1/3 Infrared ST1230, Campina Grande do Sul, Paraná, Brazil). The recorded images of each bird were analyzed at the same time by one evaluator. Every ten minutes, the activities of the animals were recorded following the focal sampling technique (Altmann, 1974) in a pre-defined ethogram (Figure 1) elaborated for spectacled parrot (*Amazona pretrei*) of the order *Psittaciformes* in captivity (Prestes, 2000). With the total number of registered behaviors for each bird, the percentage of each behavior categories was calculated for each experimental unit.

The data were evaluated by ANCOVA after the normality test using Shapiro-Wilk test and the means were compared by F test. The values of each experimental unit obtained during the first period of experiment (1-4 days) were used as covariate. For the variables that not achieved the normality, Kruskal-Wallis test was used. In all cases, $\alpha = 0.05$. All statistical analysis was performed using the Action 2.3 statistical program.

RESULTS AND DISCUSSION

The room temperature was $35.0 \pm 2.0^\circ\text{C}$ during the daylight (6h00min to 18h00min) and $25.0 \pm 2.0^\circ\text{C}$ during the dark (18h01min to 5h59min). No interaction or effect of enriched environment or period of evaluation was observed ($P > 0.15$) in the body surface temperature of cockatiels (Table 1). The birds, warm-blooded animals, control body temperature through behavioral changes or else physiological mechanisms, such as increase or decrease of the blood flow in the body surface which, in turn, facilitates or difficult the body heat dissipation to the environment when temperatures are outside the comfort zone (Macari *et al.*, 1994). Thus, the temperature of the body surface of the birds can be used to evaluate the physiological response of the animal to an environment that is not conducive to their welfare (Nascimento *et al.*, 2014).

The enriched environment with collard green stalks increased ($P < 0.01$) the feed intake of commercial diet but not ($P > 0.05$) the seed mix after the use of enrichment. The approximate consumption of collard green stalks during the experimental period was 180 g. The intake of the stalks shows that the birds interacted with the material. It is known that foraging is an important feature for cockatiels, which, in their natural habitat, can take up to 70% of its activity time (Péron and Grosset, 2014). In this case, the use of substances that enhance this natural behavior of captive birds is important because it promotes not only animal welfare but also the captive breeding practice and possible reintroduction of the birds to the natural environment (Coulton *et al.*, 1997).

Use of collard green...

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Behavioral Observation Activities and Environmental Enrichment

Date: _____ Schedule: _____ Responsible: _____ Cage / Species: _____

Ethogram: MP-moving laterally on the perch; WS-walking on the screen cage; FW-flapping the wings; SB-standing on the belly; SP-standing on the perch; SS-stopping on the screen; SF-stopping on the cage floor; CF-clearing feathers with its beak; WP-wagging its plumage; UD-perching upon the drinker or drinking water; UF-perching upon the feeder or eating; BR-biting the roost; BP-biting the pot supply; BS-biting the screen cage.

Time	Behavioral categories													
	Locomotion			Rest				Maintenance		Feeding		Undesirable activities		
	MP	WS	FW	SB	SP	SS	SF	CF	WP	UD	UF	BR	BP	BS
10														
20														
30														
40														
50														
60														

Description of behavioral activities:

MP-Moving laterally on the perch: the bird moves laterally on the perch.
 WS-Walking on the screen cage: the bird moves on the screen of the cage, both on the floor and on the side.
 FW-Flapping wings: the bird flaps its wings several times without shaking its plumage.
 SB-Standing on the belly: the bird rests, standing with its legs bent, its weight on its belly, and its claws and neck fully retracted over its chest.
 SP-Standing on the perch: when on the perch in a neutral posture, the bird keeps its legs slightly apart. The feathers are not ruffled, and the neck is in a normal position.
 SS-Stopping on the screen: the bird remains on the screen of the cage, showing no movement.
 SF-Stopping on the floor of the cage: the bird is standing on the cage floor, showing no movement.
 CF-Clearing feathers with its beak: the bird uses its beak to clean its feathers. This act can be performed either at the tip or near the belly, with its head toward the back. The bird bends its body corresponding to the wing.
 WP-Wagging its plumage: the bird's feathers bristle on the body, especially the neck, chest, and back, accommodating penalties after two or three jolts throughout the body.
 UD-Perching upon the drinker or drinking water: the bird perches on the pot of water or drinks water from the pot's nozzle.
 UF-Perching upon the feeder or eating: the bird remains on the feeder, stopping or eating.
 BP-Biting the perch: the bird tries to destroy the roost cage by biting it with its beak.
 BD-Biting the drinker of feeder: The bird tries to destroy the pot supply by gnawing it with its beak.
 BS-Biting the screen cage: the bird tries to destroy the screen cage by biting it with its beak.

Figure 1. Ethogram for evaluating the behavioral parameters of individual cockatiels (*Nymphicus hollandicus*).

Table 1. Feed intake and body surface temperatures of captivity cockatiels (*Nymphicus hollandicus*) during and after the use of collard green stalks as environmental enrichment in the cages (n = 8)

Item	Body surface temperature (°C)			Feed intake (g/bird/day)	
	Maximum	Average	Minimum	Commercial diet	Seed mix
Without enrichment					
During	33.8	30.5	28.7	2.2	10.7
After	33.1	30.3	28.5	2.3	10.6
With enrichment					
During	34.3	31.2	29.0	3.3	10.9
After	33.9	31.0	28.9	3.9	11.6
CV (%)	1.70	1.35	0.95	18.5	16.6
P value					
Enrichment (E)	0.15	0.36	0.26	<0.01	0.24
Evaluation period (PA)	0.45	0.62	0.52	0.79	0.14
E x PA	0.25	0.66	0.46	0.74	0.80

CV (%) - coefficient of variation

Also in relation to nutritional balance, it is known that the cockatiel is a small granivorous bird, which can survive without drinking large amounts of water, metabolically producing enough water through the oxidation of carbohydrates and fats (MacMillen and Baudinette, 1993). In this case, collard green stalks can be used not only as environmental enrichment, but also as an indirect source of water for parrots, since vegetables and fresh vegetables have approximately 85% water in their composition (Koutsos *et al.*, 2001).

During the use of collard green stalks as environmental enrichment, lower sleeping activities were registered ($P < 0.05$) in the enriched group (Table 2). Similarly, in blue-fronted parrot (*Amazona aestiva*, Psittacidae) it was observed that the rates of stationary display of inactive behavior (sleep) were significantly lower during the enrichment phase and significantly increased post enrichment (Andrade and Azevedo, 2011). The result obtained in the present study suggests that collard green aroused

the interest or curiosity of the birds without to affect significantly other specific behavioral characteristics. To reduce the 'sleep' activity indicates reduction of idleness which could contribute to reduce undesirable activities (Carlstead and Shepherdson, 2000).

After removal of enrichment, there is a numerical tendency towards increase in bird undesirable activities such as biting the screen cage. In general, environmental enrichment involves several activities like searching for food, social interactions, training or intellectual stimulation and physical activities (Meehan *et al.*, 2003) which can substantially contribute to the psychological well-being of the birds (Jones, 2007). This study shows that birds that once lived on favorable terms and are removed from their environments can manifest undesirable behavior. In such cases, the use of collard greens stalks can mitigate the problems arising from the marketing of cockatiels or seizure of wildlife birds kept in illegal captivity.

Table 2. Behavioral expression of captivity cockatiels (*Nymphicus hollandicus*) during and after the use of collard green stalks as environmental enrichment in the cages (n = 8)

Behavioral categories	Without enrichment		With enrichment		P =
	During	After	During	After	
Locomotion activities, %					
Lateral displacement of the perch	0.36	0.18	0.36	0.18	0.80
Walking on the screen cage	3.26	3.26	4.17	2.36	0.79
Maintenance activities, %					
Shaking plumage	-	-	-	-	-
Cleaning feathers	8.88	8.52	5.98	8.70	0.40
Resting activities, %					
Sleeping	28.44 a	36.78 a	11.78 b	36.05 a	<0.01
Standing on the perch	15.94	8.88	7.25	6.89	0.10
Meadow on the screen	3.44	1.99	5.25	2.36	0.89
Standing on the ground	16.12	17.21	14.13	11.05	0.11
Feeding activities, %					
Ingesting water	3.99	3.99	3.26	2.54	0.92
Ingesting food	8.88	12.32	9.60	15.76	0.12
Undesirable activities, %					
Biting the roost	0.18	0.36	0.00	0.18	0.82
Biting the pot	-	-	-	-	-
Biting the screen cage	1.99	6.34	2.72	13.59	0.06

*Means followed by the same letter do not differ by test Kruskal-Walis ($P < 0.05$).

No effect of use of collard green stalks was observed ($P > 0.10$) in the others behavior characteristics of the birds. These results are consistent with those obtained for superficial

body temperature of birds. Behavior is generally considered to be the most effective form of body temperature regulation (Schlader *et al.*, 2011). Increase or decrease of body surface temperature

in birds maintained in the same conditions could indicate higher or lower intensity of heaters movements (Refinetti *et al.*, 1989) or indicate presence of acute stress (Olivier *et al.*, 2003). Acute stress or stress-induced hyperthermia is mediated by the autonomic nervous system and occurs before and during exposure to anxiogenic stimuli or stress inducers such as noise, heat, handling, novelty or pain (Kleitman, 1945; Renbourn, 1960; Bermant *et al.*, 1979; Briese, 1995; Briese and DeQuijada, 1970; Marazziti *et al.*, 1992). Thus, it is possible to presume that collard green stalks did not increase heating movements and consequently did not affect the bird's thermal status.

Although environmental enrichment is a potentially promising strategy to reduce fear responses of captive parrots and similar birds, little is known about the elements necessary for effective environmental enrichment for these birds. Thus, it is essential to select carefully the environmental enrichment used and adjust the complexity of the environment to the behavioral characteristics and ability of each species to interact with the enrichment item.

Behavioral studies with cockatiels (*N. hollandicus*) kept in captivity are scarce in the literature. Most studies with psittacines were based on parrots. In the present study, the use of cabbage stalks as environmental enrichment was not sufficient to alter significantly the behavior of the birds; however, it cannot be said that the use of enrichment was not effective. We observed that all birds interacted with the enrichment item immediately so that we can affirm that this substrate can be used to enhance the ambience conditions of cockatiels kept in captivity.

CONCLUSION

Collard green stalks can be used as environmental enrichment, forming part of food preferences of cockatiels. However, its use does not expressively alter the behavior of birds kept in captivity.

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