FEAR LEVEL AND BEHAVIOUR OF GROWING RABBITS HOUSED IN INDIVIDUAL, BICELLULAR AND COLLECTIVE CAGES

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ABSTRACT

During growth (from 27 to 75 d of age), 384 rabbits were kept into 72 individual cages (72 rabbits), 48 bicellular cages (2 rabbits/cage; 96 rabbits) and 24 collective cages (9 rabbits/cage; 216 rabbits). To evaluate the effect of the housing system on fear level and behavioural pattern of rabbits at two ages (39-45 d and 66-73 d of age), the tonic immobility test and the open field test were performed and behaviour was video-recorded. At the tonic immobility test, the number of attempts to induce immobility was lower (1.38) and the duration of immobility was higher (47.8 sec) in rabbits from individual cages than in those from bicellular (1.72 number of attempts and 25 sec of immobility) and collective cages (1.99 number of attempts and 25.0 sec of immobility) ($0.05<P<0.01$). During the open field test, rabbits from individual and bicellular cages showed higher latency (38.8 and 40.3 vs. 27.0 sec), lower number of total (73.3, 81.7 and 91.9) and central displacements (3.6 and 2.8 vs. 5.4), ran for a shorter time (11.8 and 13.6 sec vs. 17.7 sec) and bit less some parts of the pen (5.5 and 9.1 sec vs. 28.2 sec) compared to rabbits kept in collective cages. During the 24 hours, rabbits in individual and bicellular cages spent less time for allo-grooming (0.34% and 0.19% vs. 1.44%), moving (0.74% and 0.60% vs. 1.32%) and running (0.08% and 0.03% vs. 0.21%) than rabbits in collective cages ($0.01<P<0.001$). The lowest numbers of rearing and hops were observed in rabbits kept in bicellular cages. In conclusions, rabbits in individual cages exhibited the highest fear level, a reactive coping strategy and an incomplete behavioural pattern; rabbits housed in collective cages showed the lowest fear levels, had proactive coping strategy and the possibility of expressing more behaviours; rabbits in bicellular cages exhibited a not consistent pattern of fear in the tonic immobility and open field tests and a less defined coping strategies. These rabbits were likely in a less stressful condition compared to animals in individual cages since locomotory possibilities were even more limited, but social contacts were allowed.

Key words: Housing system, fear level, behaviour, growing rabbits.

INTRODUCTION

Because of their recent domestication, rabbits exhibit several wild behaviours (Trocino and Xiccato, 2006; Verga *et al*., 2007) and their welfare may be somewhat challenged under the conditions of commercial intensive rearing (Szendrő and Dalle Zotte, 2011). In some European Countries (e.g. Italy), the housing conditions of fattening rabbits in small-size bicellular cages (2 rabbits/cage) do not permit rabbits to express fully their typical activities and social behaviour; rabbits may be bored, spend most of their time resting and show some stereotypes, like biting or licking the cage (Podberscek *et al*., 1991; Szendro and Dalle Zotte, 2011). Under these stressful conditions, fear and anxiety of animals may increase (Forkman *et al*., 2007) and, thus, their physiology and immune reactions impair (Koolhaas *et al*., 1999). Few experimental tools and data are available that measure fear level in rabbits: the tonic immobility test has been used to evaluate their fear towards humans (Ferrante *et al*., 1992; Trocino *et al*., 2004 and 2008; Verwer *et al*., 2009) and the open field test has been performed to get information on their fear towards an unknown environment (Meijsser *et al*., 1989; Ferrante *et al*.,

1992; Xiccato *et al*., 1999). The present study aimed at evaluating whether housing in individual, bicellular and collective cages may affect fear level and behavioural pattern of growing rabbits.

MATERIALS AND METHODS

Animals, experimental groups and behavioural recordings

Three hundred eighty-four hybrid rabbits of both genders were reared from weaning, at 27 days of age, until 75 days. The animals were divided into three experimental groups as follows: 72 rabbits were put into individual cages (25 x 40 x 30 cm; 1000 cm^2 /rabbit; 10 rabbits/m^2); 96 rabbits were put into 48 bicellular cages (2 rabbits/cage) (28 x 40 x 30 cm; 560 cm^2 /rabbit; 18 rabbits/m^2) and 216 rabbits were put into 24 open-top collective cages (9 rabbits/cage) (100 x 50 cm, 555 cm^2 /rabbit; 18 rabbits/m^2). The animals were controlled for performance and slaughter results as described by Trocino *et al*. (2011).

The tonic immobility test was performed on 48 rabbits (16 rabbits per housing system) at 42 days of age and repeated at 70 days on the same animals. The test was performed in the same barn where individual, bicellular and collective cages were placed. The operator took the rabbit out of its cage and induced immobility by turning the animal on its back as described by Ferrante *et al*. (1992). The immobile rabbit was laid down on its back on a V-shape wooden structure. A maximum of three attempts was performed to induce immobility and the rabbit was left in the immobility condition no more than 180 seconds.

The open field test was performed on 48 rabbits (16 rabbits per housing system) at 39 days of age and repeated on the same animals at 73 days. A pen (2 x 2 m) with 0.80 m-high wooden walls and a plastic floor divided into nine numbered squares was used (Meijsser *et al*., 1989; Ferrante *et al*., 1992). The total duration of the test was 12 minutes per animal: each rabbit was taken out of its cage and put in a closed small box (21 x 30 x 25 cm) connected to the pen by a sliding door; after one minute, the sliding door was opened permitting the animal to enter into the pen; if after one minute the rabbit was still in the small box, it was gently pushed into the pen, the sliding door was closed and the behaviour of the rabbit was recorded during 10 minutes. The following behaviours were considered: total and central displacements (number of squares crossed in the pen and in the middle of the pen), movement, running, exploration, hops, standing still (time spent still with fore and hind legs not stretched and on the ground), rearing (number of times the rabbit upheaves on its hind legs); self-grooming; digging, biting; resting (Ferrante *et al*., 1992).

Rabbit behaviour was video-recorded on 48 individual cages, 24 bicellular cages and 8 collective cages, corresponding to the observation of a total of 168 rabbits. The video recording was performed during 24 hours at two ages, 45 days of age and repeated at 66 days on the same cages, and using the "scan sampling" method, that is one minute was recorded each half an hour per each cage. During the night a minimal light was used (15 W) to avoid disturbance to nictameral activities of rabbits. The following behaviours were analysed: resting (with crouched or stretched body), self-grooming, allo-grooming, feeding, drinking, moving, running, hopping, standing still, rearing, biting, sniffing, abnormal behaviours and aggressive interactions (Morisse *et al*., 1999; Dal Bosco *et al*., 2002).

Statistical analysis

Data of reactivity and behaviour were tested for a normal distribution by using the Shapiro-Wilk statistic. Normally distributed data were analysed by ANOVA with housing system and age as the main effect and using the GLM SAS procedure (Statistical Analysis System Institute, Inc., NC, USA). Not-normally distributed data were submitted to non parametric analyses of variance (Kruskal-Wallis and Mann-Whitney tests) using the NPAR1WAY SAS procedure. The Bonferroni *t* test was used to compare means by group of housing systems. Differences among means with $P<0.05$ were accepted as representing statistically significant differences. Differences among means with $0.05<P<0.10$ were accepted as representing tendencies to differences. The values are reported on table as the average of

the two ages at different tests and video recording. The effect of age is not reported on tables nor discussed.

RESULTS AND DISCUSSION

The tonic immobility test mimes a predator attack to which the animal may react through struggle or immobility and, with certitude in birds, the duration of immobility is positively correlated with the fear level (Forkman *et al*., 2007). At this test, the percentage of rabbits that entered into immobility within three attempts (sensible) tended to be higher in rabbits in individual cages compared to those in bicellular and collective cages (90.6% *vs.* 75.0 and 68.8%; P<0.10) (Table 1). The number of attempts to induce immobility in sensible rabbits was lower and the duration of immobility was higher in rabbits from individual cages than in those from bicellular and collective cages (0.05<P<0.01).

Table 1: Response of rabbits to the tonic immobility test (average of recordings at 42 and 70 days of age)

	Cage type			
	Individual	Bicellular	Collective	Probability
Records, n	32	32	32	
Sensible rabbits[1,2] (%)	90.6	75.0	68.8	0.09
Attempts, n	1.38[A]	1.72[B]	1.99[B]	**
Immobility, sec	47.8[b]	25.0[a]	25.0[a]	*

n.s., non significant; *P<0.05, **P<0.01. [1]Rabbits that underwent into immobility within three attempts. [2]Probability of χ^2 test

During the open field test, rabbits in collective cages were more bold and active compared to rabbits kept in individual or bicellular cages (Table 2): a higher percentage of these rabbits entered spontaneously into the arena; they showed the highest number of total and central displacements; they spent more time running and biting some parts of the pen. However, similarly to what described in birds and sheep, social isolation during the test could condition the behaviour of rabbits from collective cages, since movement may be related to group reinstatement purpose, besides curiosity (Forkman *et al*., 2007).

One the base of our results on fear tests and according to Khoolhaas *et al*. (1999), we could argue that the housing system affected the copying strategies of rabbits towards stressors and environment: rabbits in individual cage were "reactive", that is they were less prone to face the stressors and rather attempted to escape them through immobility; rabbits from collective cages were "proactive", that is they reacted to the stressors with aggressive and locomotion behaviours. Rabbits in bicellular cages had an ambiguous coping strategy, that is they were proactive during the tonic immobility test and reactive during the open field test.

As what concerns the behavioural pattern (Table 3), rabbits spent most of their time in resting (67%), allo grooming (16%), feeding and drinking (13%) which confirms previous studies (Morisse and Maurice, 1997; Morisse *et al*., 1999; Martrenchar *et al*., 2001; Dal Bosco *et al*., 2002; Postollec *et al*. 2006 and 2008; Trocino *et al*., 2008). The housing system did not affect the expression and the amount of the main activities (resting and feeding); stereotypic behaviours did not occur, but the decrease in self-grooming and the increase in allo-grooming in collective cages compared to individual and bicellular cages may be positively considered. Rabbits in individual and bicellular cages spent less time allo-grooming, moving and running than rabbits in collective cages (0.01<P<0.001). The numbers of rearing and hops were significantly different according to the housing system with the lowest occurrence in rabbits kept in bicellular cages. In fact, in individual, bicellular cages or small conventional collective cages with 4-6 animals, rabbits cannot move or run, cannot hop or rearing, especially at older ages, as observed both in the present and in previous trials (Podberscek *et al*., 1991; Dal Bosco *et al*., 2002; Postollec *et al*., 2006).

Table 2: Behaviour during the open field test (average of recordings at 39 and 73 days of age).

	Cage type			
	Individual	Bicellular	Collective	Probability
Records, n	32	32	32	
Entered animals[1,2], %	56.2	46.8	81.2	**
Total displacements[3], n	73.3^{a}	81.7^{ab}	91.9^{b}	*
Central displacements, n	3.6^{ab}	2.8^{a}	5.4^{b}	*
Exploration[3], sec	463	451	441	0.07
Movement, sec	65.5	65.9	66.9	n.s.
Running, sec	11.8^{a}	13.6^{ab}	17.7^{b}	*
Standing still, sec	46.1	50.2	32.0	n.s.
Biting, sec	5.5^{A}	9.1^{A}	28.2^{B}	***
Digging, sec	2.0	2.4	3.5	n.s.
Resting, sec	1.8	3.2	0.7	n.s.
Hops, n	4.1	3.7	4.8	n.s.

n.s., non significant; *P<0.05, **P<0.01, ***P<0.001.
[1]Rabbits which entered the pen spontaneously within 60 seconds. [2]Probability of χ^2 test. [3]Data with normal distribution.

Table 3: Behaviour (% of observations) of rabbits during 24 hours: effect of the housing system and of the age of rabbits (average of recordings at 45 and 66 days of age).

	Cage type			
	Individual	Bicellular	Collective	Probability
Records, n.	96	96	144	
Resting[1], %	63.9	67.7	67.9	n.s.
Feeding[1], %	12.1	11.6	8.7	n.s.
Drinking, %	1.30^{A}	2.24^{B}	2.35^{B}	***
Self-grooming[1], %	18.4	15.0	14.8	n.s.
Allo-grooming, %	0.34	0.19	1.44	***
Sniffing, %	1.77	1.54	2.16	n.s.
Movement, %	0.74^{A}	0.60^{A}	1.32^{B}	**
Running, %	0.08^{A}	0.03^{A}	0.21^{B}	***
Standing still, %	1.03	0.97	1.09	n.s.
Biting, %	0.27	0.11	0.00	n.s.
Rearing, n	0.58^{B}	0.13^{A}	0.44^{B}	***
Hops, n	0.79^{b}	0.56^{a}	0.86^{b}	*

n.s., non significant; *P<0.05, **P<0.01, ***P<0.001. [1]Data with normal distribution.

CONCLUSIONS

In the condition of the present trial, rabbits in individual cages exhibited the highest fear level, a reactive coping strategy and an incomplete behavioural pattern. The limited space available for some activities and the absence of congeners were likely to increase the stress of rabbits and severely threatened their welfare. On the contrary, rabbits housed in collective cages with nine animals showed the lowest fear levels, had proactive coping strategy and the possibility of expressing more behaviours. Finally, rabbits in bicellular cages exhibited less clear coping strategies. Despite movement possibilities were even more limited compared to animals in individual cages, rabbits kept in bicellular cages likely took advantage of social contacts with the cage companion.

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