

## **EFFECT OF GROUPING TIME AND GROUP STABILITY ON BEHAVIOR AND AGGRESSION AMONG RABBIT DOES IN A PART-TIME HOUSING SYSTEM**

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### **ABSTRACT**

To evaluate aggression and behaviour of reproducing does in a part-time system, 48 crossbred multiparous rabbit does were housed in individual modules of a park system. A total of 12 parks with 4 does were formed by joining adjacent modules from 9 d before kindling until 2 d before kindling and from 2 d or 12 d after kindling (early or late grouping time, respectively) until weaning. Within grouping time, half of the parks maintained a stable group; the other half changed one doe every week. Behaviour in all parks was video-recorded for 24 h at 2 d, 12 d and 19 d after kindling. The number of total aggressive interactions significantly decreased from the grouping day to the 19<sup>th</sup> d of lactation ( $P < 0.001$ ), and was lower in late-grouped does compared to early-grouped ones ( $P < 0.01$ ) and in stable groups compared to variable groups ( $0.001 < P < 0.01$ ). Regarding the time spent resting, feeding, drinking and self-grooming, the observation day always had a significant effect, whereas the group stability was less relevant. Finally, at 19 d, on average, does rested individually for a 45.3% of the observation time and in contact with a mate for a 17.2% of the observation time, without significant differences according to the grouping time or the group stability ( $P > 0.05$ ).

**Key words:** lactating does, group housing, welfare, aggression, budget time.

### **INTRODUCTION**

Group housing is claimed to improve animal welfare in rabbit farms (European Parliament, 2017). Nevertheless, in the reproductive sector, continuous group housing was proved to seriously challenge welfare of reproducing does and kits and cannot be recommended (Szendrő et al., 2019). On the other hand, part-time housing systems, in which does are housed individually and in small groups for some periods, have shown some potential (Szendrő et al., 2019). Aggression and injured does after each re-grouping are an unsolved problem (Szendrő et al., 2019), however. Among the different factors affecting does aggression, the grouping time plays a key role. Late grouping after kindling is expected to decrease aggression (Zomeño et al., 2017), but a high percentage of injured does was recorded in a part-time system with late grouping (18 d after kindling) (34% at 4 d after regrouping, 53% at litter weaning; Rommers and De Greef, 2018). Moreover, since aggression is associated with hierarchy establishment, continuous aggression is likely to occur during the whole reproductive cycle under commercial conditions since new does could be introduced at any time in a group for several reasons (not pregnant, ill or dead does). Thus, the present study aimed at evaluating the effect of grouping time (early vs. late respect to kindling) and group stability (stable vs. variable) on aggression and behaviour of reproducing does at different days during the reproductive cycle after kindling.

### **MATERIALS AND METHODS**

At the 17<sup>th</sup> day of pregnancy, 48 crossbred multiparous rabbit does (3-5 kindling) were moved from a commercial farm to the experimental farm of the University of Padova and housed in individual modules of a park system (0.5 m<sup>2</sup>; 92 cm length × 53 cm width) equipped with manual feeders, automatic drinkers, and nests. A total of 12 parks was formed by removing the wire-net walls between adjacent modules to house 4

does, and their litters when present, from 9 d before kindling until 2 d before kindling and from 2 d or 12 d after kindling, in the case of the early (E) or late (L) grouping time respectively, until weaning (33 d after kindling). Within grouping time, half of the parks maintained a stable group (S) and the other half changed one doe every week (V). In the V group, every does was moved only once and the final group at the weaning time contained no more than two does belonging to the initial park/group. Litters were standardized to nine kits, controlled lactation was used, and nests were closed from 2 until 19 d after kindling. Zootechnical results are given in Birolo et al. (2018).

Behaviour of does in all parks was video-recorded with colour infrared cameras for 24 consecutive h in three moments after kindling: 2 d (after grouping for E does and changing does for the V groups); 12 d (after grouping for the L does and changing does for the V groups); and 19 d (after opening nests for all groups and changing does for the V groups). Aggressive interactions among does were recorded according to Zomeño et al. (2018) as number of biting, carousel fights, chasing, boxing, attacking, escape, and mount attempts. Also, the time spent feeding, drinking, resting (crouched or stretched body) and self-grooming was recorded. Behaviour was controlled for 30 min per h per 24 h for a total observation time of 12 h. The number of interactions among does and the time spent in the different behaviours were recorded at park level.

Data were analysed using SAS 9.4 software (SAS Institute, Cary, USA). The number of the aggressive interactions were submitted to ANOVA by applying a mixed model and the PROC GLIMMIX, using observation day (grouping day, i.e. 2 or 12 d, and 19 d after kindling), grouping time (early vs. late), group stability (stable vs. variable), observation hour and their interactions as fixed effects and the park as a random effect. A Poisson distribution was assumed for all data. Data related to time spent resting, feeding, drinking, and self-grooming were submitted to ANOVA by a mixed model and the PROC MIXED using observation day (2 d, 12 d, and 19 d after kindling), grouping time, group stability, observation hour and their interactions as fixed effects and the park as a random effect. The Bonferroni t-test was used to compare means.

## RESULTS AND DISCUSSION

The numbers of threatening, chasing, boxing, and mount attempts were very low at 12 d and null at 19 d so that no statistical analysis was possible. Regarding the main effects, the number of total aggressive interactions significantly decreased from the grouping day to the 19<sup>th</sup> d of lactation ( $P < 0.001$ ), and was lower in late-grouped does compared to early-grouped ones ( $P < 0.01$ ) and in stable compared to variable groups ( $0.001 < P < 0.01$ ) (Table 1). Nevertheless, significant interactions were recorded between observation day and grouping time as well as between observation day and group stability (Table 1). Concerning observation day  $\times$  grouping time, total aggressive interactions as well as biting, carousel-fights, and escape attempts were higher in early-grouped does than in late-grouped does, but only on the grouping day, without residual effects 19 d after kindling (i.e. 17 and 5 d after grouping for the early-grouped and late-grouped does, respectively). On the other hand, attacks were higher in late-grouped than in early-grouped does. These results confirm previous observations (Zomeño et al., 2017, 2018): does grouped only 2 d after kindling are more aggressive likely because of their stressful physiological condition compared to does grouped 12 d after kindling; thereafter, at 19 d after kindling, when does have a stable milk production and kits are free of going out from the nests, aggressive interactions among adults are reduced. Other authors (Rommers et al., 2011; Andrist et al., 2012) also observed that aggression decreased 3-5 d after grouping. Regarding observation day  $\times$  group stability, differences according to group stability were recorded at 19 d with more aggressive interactions and more escape attempts in variable groups compared to stable ones (Table 1). Differently, Andrist et al. (2012) observed more biting and boxing in variable groups only on the grouping day (after 12 d of isolation), without residual effects 3-5 days after, even if fecal glucocorticoid contents and rate of injured does were higher in does with variable group composition. Nevertheless, in the aforementioned study, variable groups only changed at regrouping time.

Regarding the budget time, the observation day always had a significant effect, whereas the group stability was less relevant (Table 2). Moreover, as for aggressive behaviors, significant interactions were found which are given in Table 2. In fact, early-grouped does spent less time resting, feeding, drinking, and self-grooming at the grouping time (2 d after kindling) than 10 d later (12 d after kindling). At 19 d after kindling, when controlled lactation ended and nests were opened, resting time increased especially with stretched body, whereas feeding and drinking time decreased compared to 2 and 12 d *post-partum*. Also late-grouped does

spent less time resting and feeding on the grouping day compared to later observations (19 d after kindling), when no difference was recorded between early-grouped and late-grouped does at 19 d. Similarly, Buijs et al. (2015) found that does spent less time resting, feeding, and grooming and more time interacting with other mates (attacking, escaping, sniffing, licking) on the grouping day (18 d after kindling) than 4 d or 12 d later. Dal Bosco et al. (2019) also showed that does in a part-time system are more active (more movements) at grouping (7 d after kindling) than during the previous isolation phase (from 4 d before until 7 d after kindling). Regarding the interactions observation day  $\times$  group stability, total resting time was not affected, but at 19 d does in stable groups spent more time resting in a stretched position compared with does in variable groups, which could be related to the higher aggression among does in this latter group. On the other hand, time spent feeding and drinking was higher in stable vs. variable groups only 12 d after kindling.

Finally, at 19 d we also checked if does were in contact or not with a mate during resting. On average, at this time, does rested individually for a 45.3% of the observation time and in contact with a mate for a 17.2% of the observation time, without significant differences according to the grouping time or the group stability ( $P>0.05$ ) (data not reported in tables).

## CONCLUSIONS

The present results confirm the great effect of the grouping time on aggression among does in a part-time system: the later the grouping with respect to kindling the lower the aggressive interactions. Maintaining a stable group does not prevent aggression at regrouping, whereas the introduction of a new doe during the reproductive cycle challenges the hierarchy within the group and can increase aggression among does. Moreover, in a part-time system, the time spent feeding, drinking, resting and self-grooming changes with the level of aggression among does and with the doe physiological status during the reproductive cycle.

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**Table 1.** Aggressive interactions and escape attempts (means of number events/observation interval<sup>1</sup>): interaction between observation day (D) (grouping day, i.e. 2 d *post-partum* in early groups or 12 d *post-partum* in late groups; 19 d) and grouping time (T) (early, E; late, L) and interaction between observation day (D) and group stability (G) (Stable, S; Variable, V).

	Observation day (D)				Observation day (D)				Prob.						RMSE	
	2/12 d		19 d		2/12 d		19 d		D	T	G	D × T	D × G	T × G		
	Grouping time (T)		Group stability (G)													
	E	L	E	L	S	V	S	V								
Observations, no.	144	144	144	144	144	144	144	144								
Pens, no.	6	6	6	6	6	6	6	6								
Total interactions <sup>2</sup>	7.65 <sup>c</sup>	2.38 <sup>b</sup>	0.37 <sup>a</sup>	0.35 <sup>a</sup>	4.94 <sup>c</sup>	5.11 <sup>c</sup>	0.04 <sup>a</sup>	0.67 <sup>b</sup>	<0.001	<0.01	<0.001	<0.01	<0.001	<0.001	<0.05	0.58
Biting	5.02 <sup>d</sup>	0.83 <sup>c</sup>	0.13 <sup>a</sup>	0.14 <sup>a</sup>	3.07 <sup>c</sup>	2.78 <sup>c</sup>	0.03 <sup>a</sup>	0.24 <sup>b</sup>	<0.001	<0.01	<0.01	<0.001	<0.001	<0.001	n.s.	0.61
Attacking	0.26 <sup>b</sup>	0.84 <sup>c</sup>	0.08 <sup>a</sup>	0.06 <sup>a</sup>	0.42 <sup>c</sup>	0.68 <sup>c</sup>	0.01 <sup>a</sup>	0.14 <sup>b</sup>	<0.001	n.s.	<0.01	<0.001	<0.001	<0.001	<0.01	0.51
Carousel-fights	0.72 <sup>c</sup>	0.27 <sup>b</sup>	0.12 <sup>ab</sup>	0.10 <sup>a</sup>	0.31 <sup>b</sup>	0.69 <sup>c</sup>	0.01 <sup>a</sup>	0.21 <sup>b</sup>	<0.001	<0.01	<0.001	<0.05	n.s.	<0.05	<0.05	0.48
Escape attempts	2.88 <sup>c</sup>	1.12 <sup>b</sup>	0.22 <sup>a</sup>	0.17 <sup>a</sup>	2.01 <sup>c</sup>	1.99 <sup>c</sup>	0.01 <sup>a</sup>	0.38 <sup>b</sup>	<0.001	<0.01	<0.001	<0.05	<0.001	n.s.	n.s.	0.35

<sup>1</sup>Observation time per day: 12 h (30 min per h per 24 h). <sup>2</sup>Total interactions = Sum of events related to biting, attacking, carousel-fights, threatening, chasing and boxing. Means of groups within the same row and interaction with different superscript letters differ significantly (Bonferroni test).

**Table 2.** Budget time (% total observation time<sup>1</sup>, means): interaction between observation day (D) (2 d *post-partum*; 12 d *post-partum*; 19 d *post-partum*) and grouping time (T) (early, E; late, L) and interaction between observation day (D) and group stability (G) (Stable, S; Variable, V).

	Observation day (D)						Observation day (D)						Prob.						RMSE
	2 d		12 d		19 d		2 d		12 d		19 d		D	T	G	D × T	D × G	T × G	
	Grouping time (T)		Group stability (G)																
	E	L <sup>2</sup>	E	L	E	L	S	V	S	V	S	V							
Observations, no.	144	-	144	144	144	144	72	72	144	144	144	144							
Pens, no.	6	-	6	6	6	6	3	3	6	6	6	6							
Resting	45.6 <sup>a</sup>	-	57.3 <sup>c</sup>	53.5 <sup>b</sup>	62.3 <sup>d</sup>	62.9 <sup>d</sup>	46.6 <sup>a</sup>	44.6 <sup>a</sup>	55.3 <sup>b</sup>	55.5 <sup>b</sup>	63.6 <sup>c</sup>	61.7 <sup>c</sup>	<0.001	n.s.	n.s.	<0.05	n.s.	n.s.	11.3
Crouched	23.1 <sup>a</sup>	-	23.6 <sup>a</sup>	37.2 <sup>b</sup>	22.6 <sup>a</sup>	20.6 <sup>a</sup>	25.0 <sup>b</sup>	21.2 <sup>ab</sup>	29.4 <sup>b</sup>	31.3 <sup>b</sup>	18.4 <sup>a</sup>	24.8 <sup>b</sup>	<0.001	<0.001	n.s.	<0.001	<0.001	n.s.	11.7
Stretched	22.5 <sup>b</sup>	-	33.7 <sup>c</sup>	16.3 <sup>a</sup>	39.7 <sup>d</sup>	42.3 <sup>d</sup>	21.6 <sup>a</sup>	23.4 <sup>ab</sup>	25.9 <sup>b</sup>	24.2 <sup>b</sup>	45.2 <sup>d</sup>	36.9 <sup>c</sup>	<0.001	<0.001	n.s.	<0.001	<0.001	<0.01	11.6
Feeding	8.12 <sup>b</sup>	-	12.3 <sup>c</sup>	11.3 <sup>c</sup>	6.86 <sup>a</sup>	5.94 <sup>a</sup>	8.36 <sup>a</sup>	7.88 <sup>a</sup>	12.7 <sup>c</sup>	10.9 <sup>b</sup>	6.39 <sup>a</sup>	6.41 <sup>a</sup>	<0.001	<0.05	n.s.	n.s.	<0.001	n.s.	4.94
Drinking	2.86 <sup>b</sup>	-	3.87 <sup>c</sup>	3.27 <sup>bc</sup>	1.86 <sup>a</sup>	1.62 <sup>a</sup>	3.15 <sup>b</sup>	2.57 <sup>b</sup>	4.04 <sup>c</sup>	3.10 <sup>b</sup>	1.67 <sup>a</sup>	1.81 <sup>a</sup>	<0.001	<0.01	<0.01	n.s.	<0.001	n.s.	2.07
Self-grooming <sup>3</sup>	11.4	-	15.3	14.0	-	-	11.0 <sup>a</sup>	11.8 <sup>a</sup>	15.4 <sup>b</sup>	14.0 <sup>b</sup>	-	-	<0.001	n.s.	n.s.	-	n.s.	<0.05	7.4

<sup>1</sup>Observation time per day: 12 h (30 min per h per 24 h). <sup>2</sup>Data of late grouped does not available at 2 d *post-partum* because still individually kept. <sup>3</sup>Recorded only at 2 and 12 d *post-partum*, thus analysed without the interaction D × T. Means of groups within the same row and interaction with different superscript letters differ significantly (Bonferroni test).