

EFFECT OF LIGHTING PROGRAMME AND NURSING METHOD ON THE PRODUCTION AND NURSING BEHAVIOUR OF RABBIT DOES

Gerencsér Zs.*, Matics Zs.[†], Nagy I.*, Radnai I.*, Szendrő É.*, Szendrő Zs.*

*Kaposvár University, Kaposvár, Guba S. str. 40, H-7400 KAPOSVÁR, Hungary.

[†]MTA - KE Research Group of Animal Breeding and Hygiene, Guba S. str. 40, H-7400 KAPOSVÁR, Hungary.

ABSTRACT: The purpose of the experiment was to analyse how the production and nursing behaviour of rabbit does are influenced by different lighting programmes and nursing methods. Rabbit does (n=119) were housed in 2 rooms. The lighting schedules were a continuous 16L:8D (16L, n=55) or an interrupted 8L:4D:8L:4D (8+8L, n=64). In both rooms, half of the does nursed their kits freely (FS, n=53), while for the other half the suckling method was changed to controlled nursing 3 d prior to the artificial insemination (AI) at day 11 (FS-CS, n=66). Lighting schedule had no significant effect on any productive trait. Seventy six percent of the 16L does nursed their kits during the dark period; however, in the 8+8L group, 50% of the nursing events occurred in the dark, 50% during the light periods, respectively. Thus the intermittent lighting could disturb the nursing behaviour of the does. The nursing method significantly affected several traits. AI/parturition, body weight of the does at kindling, number of kits born alive, litter weight at 21 d, and suckling mortality were 1.38 and 1.24 ($P<0.05$), 4.51 and 4.37 kg ($P<0.01$), 7.95 and 8.46 ($P<0.05$), 3.06 and 2.92 kg ($P<0.05$), and 5.3 and 7.3% ($P<0.05$) in the FS and FS-CS groups, respectively. Compared to the FS group, the advantage of the FS-CS group ($P<0.001$) was 16.2, 18.4, 9.3 and, 6.3% for total number of kits born, number of kits born alive, number of kits at 21 d, and total kits' weight at 21 d per AI, respectively. Due to the change in the nursing method, the frequency of multiple nursing increased. The length of the nursing period of the FS-CS group was significantly exceeded by that of the FS does. Based on these results, changing the nursing method could be considered as a possible adequate biostimulation method.

Key Words: rabbit, lighting programme, nursing method, reproductive traits, nursing behaviour.

INTRODUCTION

The reproductive performance and maternal ability of rabbit does are influenced by several factors. Alteration of the daytime by the changing seasons substantially influences the animals' productivity, primarily reproduction. This can also be observed for the European wild rabbit which is active during the night; its mating season begins in spring and ends in autumn (Lebas *et al.*, 1986).

The efficiency of artificial insemination (AI) is strongly influenced by the receptivity of the rabbit does at insemination. Inseminating at 11 d after parturition, the milk production of the does is substantial and thus their receptivity is not high enough. The proportion of receptive does, thus kindling rate, can be substantially increased by applying hormonal treatments or biostimulation methods (Theau-Clément, 2007).

The positive effects of the increased daily lighting period before AI for sexual receptivity and consequently for productivity were observed by several authors for domesticated rabbits

(Theau-Clément, 2007). When applying intermittent photoperiods compared to continuous lighting, slightly better results were found for kindling rate (Uzcategui and Johnston, 1992; Arveux and Troislouches, 1995) and litter weight (milk production; Virág *et al.*, 2000), while Hoy and Selzer (2003) found an increased frequency of twice-a-day nursing when applying intermittent photoperiods. On the contrary, no differences were found between the continuous and discontinuous lighting programmes by Theau-Clément and Mercier (2004) and Szendrő *et al.* (2004).

One of the other biostimulation techniques is changing the nursing method from free to controlled nursing 2-3 d prior to insemination. In most cases the procedure significantly increased the kindling rate and occasionally litter size as well (Bonanno *et al.*, 2004; Theau-Clément, 2007).

Although some does nurse their kits twice or three times a day, the nursing events generally occur during the night (Matics *et al.*, 2004). With the more frequent change of the dark and light periods (every 6 h), the number of daily nursing events increased (Hoy and Selzer, 2003). Moreover, applying intermittent photoperiods may increase the does' productive performance. Therefore, our hypothesis is that application of 8 h light with 4 h dark periods may have a positive effect on does' production.

The aim of the experiment was to analyse the influence of the lighting programme and nursing method on the rabbit does' production and nursing behaviour.

MATERIAL AND METHODS

The experiment was conducted at the Kaposvár University with Pannon White rabbits. The rabbits were individually housed in breeding cages at the age of 11 wk. The base area and height of the wire net cage were 84×38.5 cm (including the 26×38.5 cm sized nest box) and 35 cm, respectively. In winter the temperature ranged between 15-18°C, and it could reach 28°C in summer. All rabbits were fed *ad libitum* by the same commercial pellet throughout the whole experiment (11.0 MJ digestible energy/kg, 17.0% crude protein, 15.5% crude fibre). Water was available *ad libitum* from nipple drinkers.

The 11 wk old female rabbits were randomly housed in 2 rooms. The 2 rooms differed only in the lighting programme. In the first room, the light and dark periods were 16 and 8 h in length, respectively (16L:8D=16L; light from 6:00 to 22:00). In the second room, the light and dark periods were changed, using a rhythm of 8 h light, 4 h dark, 8 h light, 4 h dark (8L:4D:8L:4D=8+8L; light from 6:00 to 14:00 and from 18:00 to 2:00). The lighting intensity measured at the level of the rabbits ranged between 30 and 70 Lux. The experiment started at the age of 11 wk, providing enough time for the rabbits to become accustomed to the lighting regime several weeks prior to their first mating. The number of does at the beginning of the experiment was 55 and 64 in groups of 16L and 8+8L, respectively.

In both rooms, the rabbits were randomly divided into 2 subgroups according to the nursing method. In the first group, the rabbit does could freely nurse their kits from parturition till weaning (FS, n=53 does). In the second group, the nursing method was changed from free to controlled nursing 3 d prior to insemination (FS-CS, n=66 does) for biostimulation purposes (the nest boxes were open only between 8:00 and 8:30 h) and, after AI, free nursing was applied again. The FS-CS does were inseminated just after nursing.

The rabbits were first inseminated at the age of 16.5 wk; then a 42-day reproduction rhythm was applied, according to which AI occurred 11 d after kindling. The rabbits were inseminated with

diluted semen (from a single buck) and at the same time injected with 1.5 µg GnRH analogue (Ovurelin, D-Phe6-GnRH-EA, Reanal, Hungary) into their thigh muscle. Only the production of those animals which became pregnant and kindled after the first insemination was considered (n=119). The results of first kindling were not analysed. Does which failed to conceive were re-inseminated 21 d after the unsuccessful insemination.

Cross fostering was only practised within the groups. Does that died or were culled during the experiment (inadequate body condition, failed to conceive after 3 times) were not replaced. The experiment lasted for almost 1 yr and altogether data from 469 inseminations were evaluated.

Body weight of each doe was measured at kindling. Number of kits born in total, alive and dead was also recorded. Litter size and litter weight was registered 21 d after kindling, and the average individual weight was also calculated. The difference between the litter size at day 21 and number of kits after equalisation was the suckling mortality. Suckling mortality did not include the total litter loss. Total litter loss was calculated as the ratio of the number of lost litters and total number of litters (i.e. number of kindled does). Total productivity was measured by number of kits born total, number of kits born alive and number of kits and weight of kits at day 21 per AI.

After kindling, a continuous (24 h) video recording was performed for 17 d using infrared cameras (n=75 does). Time, frequency and duration of the does' nursing behaviours (length of stay in the nest box) were recorded in groups 16L (n=28) and 8+8L (n=47). In the FS and FS-CS groups, the nursing behaviour was only analysed in the 16L:8D lighting group (n=13 and n=15 does in groups of FS and FS-CS, respectively). When the effect of the nursing method was analysed, the 9-10th d of lactation were not considered because during that time controlled nursing was applied in the FS-CS group.

Data was evaluated with the SPSS 10.0 software package. Production traits (body weight of the does, number of inseminations per kindling, total number of kits born, number of kits born alive, litter size at 21 d, litter weight and average body weight of the kits at 21 d) and nursing data were analysed by means of multi-factor analysis of variance. The following model was applied:

$$Y_{ijk} = \mu + Lp_i + Nm_j + Parity_k + e_{ijk}$$

μ is the general mean,

Lp_i the effect of the lighting programme (i=16L, 8+8L),

Nm_j the effect of the nursing method (j=FS, FS-CS),

$Parity_k$ the effect of the parity order (k=parity 2, 3, 4, 5, 6, 7),

e_{ijk} is the random error.

Kindling rates, suckling mortality, distribution of nursing were analysed by chi²-test.

Average number of daily nursing events was evaluated by means of one-factor analysis of variance. The applied model was the following:

$$Y_i = \mu + Lp_i + e_i \quad \text{or} \quad Y_j = \mu + Nm_j + e_j$$

RESULTS

No significant interactions were found between the lighting programmes and nursing methods for any trait, so the effects of lighting programme and nursing method are presented separately.

Table 1: Effects of the lighting programme on the performance rabbit does.

	16L ¹		8+8L ²		SE	P-value
	No.	Mean	No.	Mean		
AI/kindling	150	1.27	210	1.32	0.03	0.237
Does' body weight at kindling, kg	146	4.47	205	4.40	0.03	0.099
Litter size						
total born	146	8.56	205	8.71	0.14	0.856
born alive	146	8.16	205	8.28	0.13	0.587
still born	146	0.40	205	0.42	0.05	0.711
at 21 d	141	7.72	200	7.82	0.07	0.759
Litter weight at 21 d, kg	141	3.01	200	2.96	0.03	0.235
Individual body weight at 21 d, g	141	393	200	385	3.67	0.248
Stillborn litters, %		2.7		2.4		0.864
Suckling mortality, %		6.3		6.5		0.846
Total litter loss, %		3.4		2.4		0.585

¹ 16L = 16L:8D. ² 8+8L = 8L:4D:8L:4D. SE: standard error.

Effect of lighting programme

The effect of the lighting schedule on the rabbit does' production is presented in Table 1. None of the productive traits were affected significantly by the lighting programme. Number of inseminations per kindling, body weight of the doe, litter size (total, alive, dead, at 21 d), litter and individual body weight at 21 d and mortality did not vary significantly with the lighting programme.

The daily number of nursing events was significantly affected by the lighting programme only between the 2nd and 4th d of lactation where the 16L does showed higher nursing frequency (Table 2). Neither during the latter stage of lactation nor between the 2nd-17th d was there any difference between the 16L and 8+8L groups. The nursing frequency distribution was independent of the lighting schedule. Differences for the frequency of once-a-day (60 and 64% for 16L and 8+8L), twice-a-day (30 and 28%) and three or four times (9 and 8%) nursing were not significant.

During lactation (between the 2nd and 17th d) in the 16L group, the proportions of once-a-day nursing did not change, those of twice-a-day decreased, while three-times-a-day increased compared to the 2nd-4th d. The opposite trend was observed in the 8+8L group until the 14th d. The proportions of once-a-day nursing decreased and those of twice-a-day nursing increased (Figure 1).

Table 2: Number of daily nursing events during lactation depending on the lighting programme.

Lactation (d)	16L ¹	8+8L ²	SE	P-value
2-4	1.43	1.24	0.04	0.008
5-8	1.39	1.46	0.04	0.375
9-10	1.20	1.26	0.04	0.456
11-14	1.55	1.59	0.04	0.593
15-17	1.61	1.41	0.05	0.051
2-17	1.45	1.42	0.02	0.418

¹ 16L=16L:8D. ² 8+8L=8L:4D:8L:4D. SE: standard error.

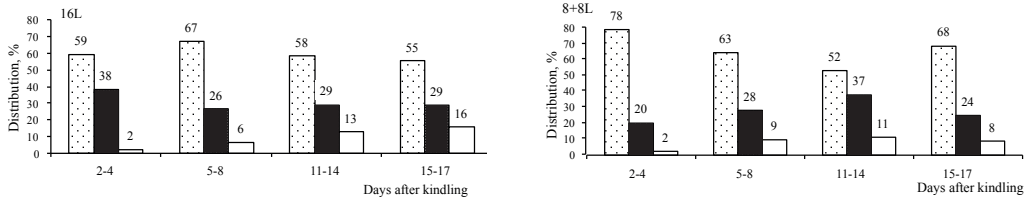


Figure 1: Occurrence rate of nursing events throughout the lactation of the 16L=16L:8D and 8+8L=8L:4D:8L:4D groups. □ Once. ■ Twice. □ Three or four times.

In the once-a-day nursing group, 76% of the 16L rabbit does nursed their kits during the dark period, showing the highest nursing frequency between 2:00 and 4:00 h (Figure 2). In the 8+8L groups, the highest nursing frequency was observed only during the first dark period, then similar proportions were recorded both in the dark and light periods (distributions of nursing event were between 2 and 12% in each 2-h period; Figure 2).

When twice-a-day nursing was recorded for the 16L group, it was observed that the first nursing event occurred almost exclusively during the dark (90.0%) and the second one during the light period (93.9%; Figure 3). Using the 8+8L lighting programme, the first nursing occurred during the first dark (between 2:00 and 6:00 h) and during the following 4 h long light (between 6:00 and 10:00 h) period (Figure 3). Defining the first dark period is, however, arbitrary; nursing between 0:00 and 2:00 h can also be considered either as an early first or as a late second nursing.

No significant differences were found for the duration of the nursing event between the 16L and 8+8L groups irrespective of the number of nursing events, except for nursing twice-a-day (1st nursing) between days 9 and 10 (Table 3). After the 2-4 d *post partum*, the duration of nursing events decreased rapidly, and then the differences were not generally different.

Effect of nursing method

Results connected to the nursing method effect are summarised in Table 4. The nursing method significantly affected several productive traits. Body weight of the FS-CS does was on average 0.14 kg lower than in the FS group. AI/kindling differed significantly, and the kindling rate of the FS-CS group was higher by 8.1% (80.6 vs. 72.5%; $P<0.05$), and litter size (total born) exceeded that of the FS group by 0.38 at level of $P<0.1$. For the number of kits born alive, the difference (0.51 kits) was significant ($P<0.05$), although at 21 d the difference was not significantly different. Litter weight at 21 d was significantly higher ($P<0.05$) in the FS group by 4.8%, but

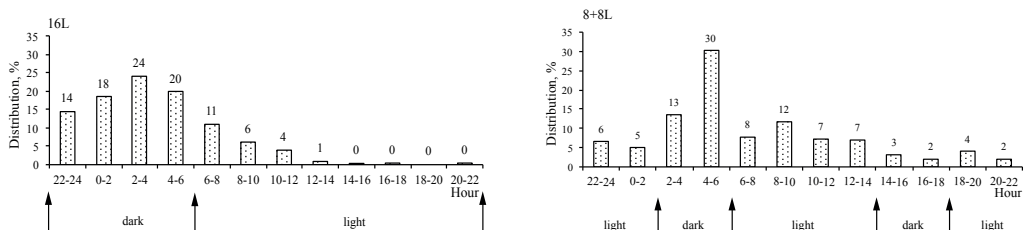


Figure 2: Daily distribution of the nursing events of the 16L and 8+8L groups for does nursing once a day. 16L=16L:8D. 8+8L=8L:4D:8L:4D.

Table 3: Duration of the nursing events (s) depending on the lighting programme.

No. of nursing events	Lactation (d)	16L ¹		8+8L ²		SE	P-value
		No.	Mean	No.	Mean		
Once	2-4	48	251 ^c	107	246 ^b	6.29	0.721
	5-8	74	184 ^b	116	176 ^a	2.59	0.131
	9-10	44	184 ^b	62	172 ^a	3.41	0.084
	11-14	64	165 ^a	95	167 ^a	2.30	0.718
	15-17	43	184 ^b	80	176 ^a	3.59	0.284
	SE			3.18		2.72	
	P-value			<0.001		<0.001	
Mean		273	191	460	190	2.07	0.732
Twice (1 st nursing)	2-4	31	267 ^b	29	262 ^c	11.5	0.845
	5-8	29	194 ^a	52	200 ^b	4.45	0.517
	9-10	11	169 ^a	15	206 ^{ab}	7.59	0.013
	11-14	32	177 ^a	69	181 ^a	3.74	0.644
	15-17	22	179 ^a	29	183 ^{ab}	5.99	0.740
	SE			5.85		4.16	
	P-value			<0.001		<0.001	
Mean		125	203	194	200	3.41	0.723
Twice (2 nd nursing)	2-4	31	240 ^b	29	245 ^c	8.67	0.774
	5-8	29	203 ^a	52	207 ^b	3.69	0.657
	9-10	11	186 ^a	15	202 ^{ab}	6.60	0.267
	11-14	32	187 ^a	69	182 ^a	3.66	0.546
	15-17	22	194 ^a	29	181 ^a	5.88	0.283
	SE			4.52		3.40	
	P-value			<0.001		<0.001	
Mean		125	205	194	199	2.72	0.319

¹ 16L = 16L:8D. ² 8+8L = 8L:4D:8L:4D. SE: Standard error.

^{a, b, c} Means within a column with different superscripts differ at $P < 0.05$.

the individual weight at 21 d did not vary with the nursing method. The FS group showed lower suckling mortality ($P < 0.05$).

Positive effects of biostimulation were clearly detected for total productivity per insemination (Table 5). Indeed, the advantage of the FS-CS group was 16.2, 18.4, 9.3 and 6.3% for total

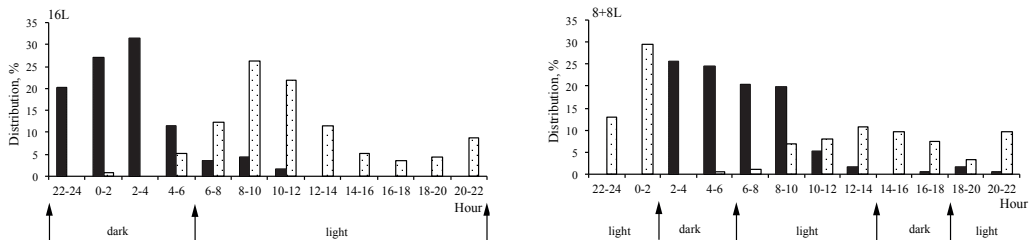


Figure 3: Daily distribution of the nursing events of the 16L=16L:8D and 8+8L=8L:4D:8L:4D groups for does nursing twice a day. ■ First nursing. ▨ Second nursing.

Table 4: Effects of the nursing method on the rabbit does' production.

	FS ¹		FS-CS ²		SE	P-value
	No.	Mean	No.	Mean		
AI/kindling	160	1.38	200	1.24	0.03	0.020
Does' body weight at kindling, kg	158	4.51	193	4.37	0.03	0.007
Litter size						
total born	158	8.44	193	8.82	0.14	0.096
born alive	158	7.95	193	8.46	0.13	0.047
still born	158	0.49	193	0.36	0.05	0.666
at 21 d	156	7.85	185	7.72	0.07	0.688
Litter weight at 21 d, kg	156	3.06	185	2.92	0.03	0.012
Individual body weight at 21 d, g	156	395	185	382	3.67	0.067
Stillborn litters, %		1.3		3.5		0.175
Suckling mortality, %		5.3		7.3		0.036
Total litter loss, %		1.3		4.2		0.107

¹FS=free nursing. ²FS-CS=free nursing was changed to controlled nursing 3 d before AI. SE: standard error.

number of kits born, number of kits born alive, number of kits alive at 21 d and litter weight at 21 d, respectively, compared to the FS group ($P<0.001$).

Changing the nursing method had no effect on the average number of daily nursing events (Table 6). The higher nursing frequency of the FS-CS group after the 11th d was not significantly different from that of does in the FS group.

Prior to the change of nursing method (1-8th d of lactation), the occurrence rate of once-a-day and twice-a-day nursing was significantly different between the FS and FS-CS groups ($P=0.039$ and $P=0.012$, respectively). The distribution of nursing 3 and 4 times a day was the same in both groups ($P=0.415$, Figure 4). After using controlled nursing, at the 9th-10th d of lactation (between the 11th and 17th d) multiple nursing occurred in a higher frequency in the FS-CS group compared to the FS rabbit does, but differences were not significant in does nursing once or twice a day. Compared to the period when the nursing method was not yet changed (between days 1-8 and 11-17), the occurrence rate of once-a-day nursing significantly decreased by 17% ($P=0.015$), whereas for 3 and 4 times a day it significantly increased by 12% ($P=0.008$, Figure 4).

Table 5: Effect of the nursing methods on the productivity per insemination.

	FS ¹		FS-CS ²		SE	P-value
	No.	Mean	No.	Mean		
No. of kindling		0.725		0.806		0.020
No. of kits						
born total	158	6.12	193	7.11	0.11	<0.001
born alive	158	5.76	193	6.82	0.10	<0.001
at 21 d	156	5.69	185	6.22	0.05	<0.001
Litter weight at 21 d, kg	156	2.21	185	2.35	0.02	0.001

¹FS = free nursing. ²FS-CS = free nursing was changed to controlled nursing 3 d before AI. SE: standard error.

Table 6: Number of the nursing events during lactation depending on the nursing method.

Lactation (d)	FS ²	FS-CS ³	SE	<i>P</i> -value
2-4	1.46	1.41	0.06	0.682
5-8	1.48	1.31	0.06	0.142
9-10 ¹	1.44	1.00	-	-
11-14	1.45	1.63	0.07	0.197
15-17	1.54	1.66	0.09	0.506
2-17	1.49	1.51	0.03	0.688

¹ The period of changing the nursing method (between the 9th and 10th d) was not included in the total evaluated period (between the 2nd and 17th d). ² FS=free nursing. ³ FS-CS=free nursing was changed to controlled nursing 3 d before AI.

In the FS group, during the first 8 d and between 11-17th d of lactation 76 and 88% of once-a-day nursing occurred between 22:00 and 6:00 h, respectively (Figure 5a). In the FS-CS group, during the first 8 days of lactation the distribution of the nursing events across the different parts of the day was similar to that of the FS group: 80% of the nursing was recorded between 22:00 and 6:00 h (Figure 5b). Between the 11th and 17th d of lactation (after changing the nursing method), this ratio decreased and only 62% of the does nursed their kits between 22:00 and 6:00 h. Thirty nine percent of the daily nursing events were observed between 6:00 and 12:00 h.

For twice-a-day nursing in the FS groups, between the 1st-8th d of lactation the first and second nursing almost exclusively occurred during the dark (97%) and during the light (100%) period, respectively (Figure 6a). There was a 6-h long period between the 2 peaks of the nursing events. Between the 11th and 17th d of lactation, the first and second nursing events occurred in a wider period and were less markedly differentiated than between the 1st and 8th d of lactation. The first and second nursing events of the FS-CS group were recorded most frequently between 22:00 and 4:00 h and between 6:00 and 12:00 h, respectively. During the 11-17th d of lactation, the time of nursing events occurred in a wider period, so the first and second nursing events also partly coincided in this group (Figure 6b).

Throughout lactation the FS-CS does had longer nursing times irrespective of the number of daily nursing events (Table 7). Significant differences were found for the mean duration of the nursing event, but in certain lactation stages the differences were also statistically proven.

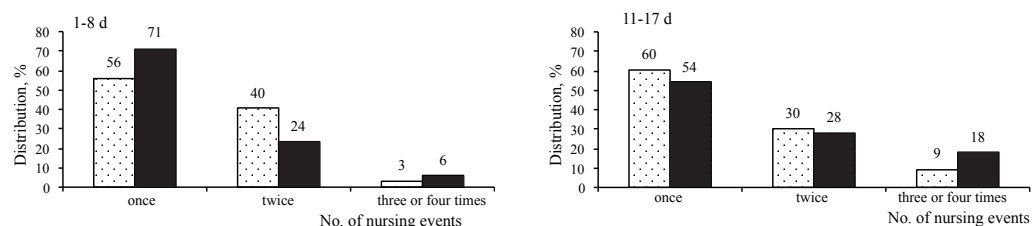


Figure 4: Number of daily nursing events before (between the 1st and 8th d of lactation) and after changing the nursing method (between the 11th and 17th d of lactation) of the FS and FS-CS groups. □ FS=free nursing throughout the lactation, ■ FS-CS= free nursing except for the last 3 d prior to AI when the does can nurse their kits once a day.

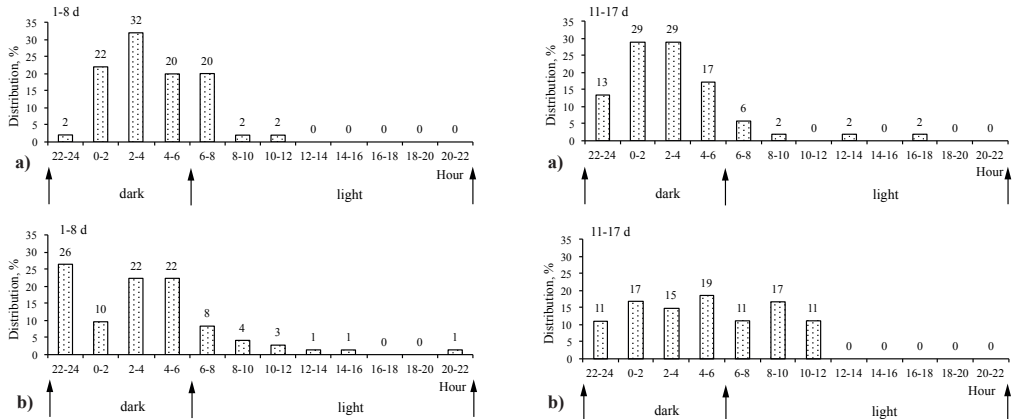


Figure 5: Daily distribution of the nursing events of the a) FS and b) FS-CS does nursing once a day during the first 8 d and between the 11th and 17th d of lactation. FS=free nursing, FS-CS=free nursing was changed to controlled nursing 3 d before AI.

DISCUSSION

Effect of lighting programme

Based on previous results, it could be expected that using intermittent lighting might have positive effects on the rabbits' production. In natural mating, Arveux and Troislouches (1995) observed that applying a 8L:4D:8L:4D lighting programme increased pregnancy rate by 15 points of percentage (67.6 vs. 82.6%), litter size (11.6 vs. 12.3) and decreased kits' mortality before weaning (6.1 vs. 1.4%). Uzcategui and Johnston (1992) carried out an experiment with Rex rabbits. When various intermittent lighting programmes were used (1L:3.5D:1L:3.5D:1L:14D; 1L:4.5D:1L:4.5D:1L:12D; 1L:5.5D:1L:5.5D:1L:10D), the authors found that the pregnancy

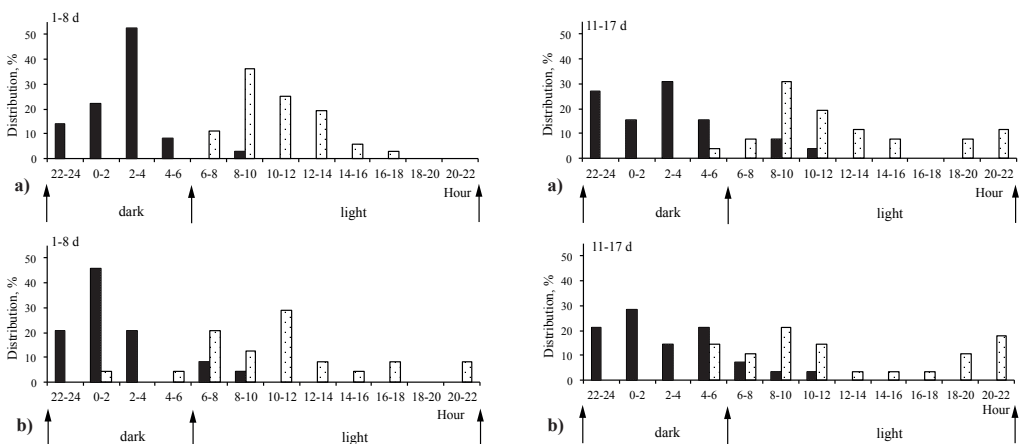


Figure 6: Daily distribution of the nursing events of the a) FS and b) FS-CS does nursing twice a day during the first 8 d and between the 11th and 17th d of lactation. FS=free nursing, FS-CS=free nursing was changed to controlled nursing 3 d before AI. ■ First nursing. □ Second nursing.

Table 7: Duration of the nursing events (s) depending on the number of daily nursing events, time of lactation and nursing methods.

No. of nursing events	Lactation (d)	FS ¹		FS-CS ²		SE	P-value
		No.	Mean	No.	Mean		
Once	2-4	20	247	28	254	10.2	0.748
	5-8	30	167	44	196	4.34	0.001
	9-10	14	177	30	187	5.38	0.407
	11-14	32	161	32	169	3.48	0.222
	15-17	21	174	22	194	6.95	0.156
	Mean	117	181	156	199	3.18	0.007
Twice (1 st nursing)	2-4	17	250	14	287	16.8	0.281
	5-8	19	177	10	226	6.76	<0.001
	9-10	11	169			8.76	
	11-14	15	172	17	181	4.54	0.356
	15-17	11	173	11	184	7.42	0.491
	Mean	73	191	52	219	5.85	0.020
Twice (2 nd nursing)	2-4	17	217	14	268	12.8	0.046
	5-8	19	196	10	218	6.15	0.087
	9-10	11	186			8.35	
	11-14	15	168	17	204	5.85	0.001
	15-17	11	186	11	201	9.10	0.415
	Mean	73	192	52	223	4.52	0.001

¹ FS = free nursing. ² FS-CS = free nursing was changed to controlled nursing 3 d before AI. SE: standard error.

rate improved by 9.1% (64.7 vs. 55.6%). However, the differences in these studies were not significant. Applying a lighting programme of 1.5L:4D:1.5L:4D:1L:12D, Virág *et al.* (2000) found 23% difference for milk yield (4.68 vs. 3.80 kg milk, $P < 0.1$). Hoy and Selzer (2003), using a lighting programme of 6L:6D:6L:6D, observed 2 daily peaks of nursing behaviour. According to Hoy and Selzer (2003), the light-dark change in the photoperiod is a “timer” for nursing activity. Based on the findings of these studies, improvement of both reproductive performance and milk yield was to be expected. However, compared to a continuous lighting programme, Theau-Clément and Mercier (2004) and Szendrő *et al.* (2004) found no differences for any trait when an 8L:4D:8L:4D lighting programme was applied. No significant differences were found for any trait by the present study either. Comparing our results with those of different studies, it has to be noted that although Arveux and Troislouches (1995) applied the same lighting programme as in this study, they nevertheless used natural mating. In natural mating, the receptivity has less importance because the mating only concerns receptive does which accept the males’ advances. Others (Uzcategui and Johnston, 1995; Virág *et al.*, 2000) applied different lighting programmes compared to those in our study. In both studies, the total length of the longer dark period per day was the same in the experimental and control groups and intermittent lighting was only performed in the part of the day that corresponded to the light period of the control group. In contrast, using an 8L:4D:8L:4D lighting schedule the day is divided into two light and dark periods of equal length.

In terms of the whole lactation, no differences were found between the 16L and 8+8L groups for the number of daily nursing events or for the proportion of rabbit does showing once-a-day,

twice-a-day or multiple daily nursing. These findings did not concur with those of Hoy and Selzer (2003), which may be explained by the different intermittent schedule (6L:6D:6L:6D). However, in the present study the occurrence rate of twice-a-day or multiple daily nursing was more than doubled between the 2nd-4th d and 11-14th d of lactation in the 8+8L (Figure 1), although in relation to the whole lactation period no significant differences were observed. YoungLai *et al.* (1986) analysed the melatonin production depending on the photoperiod (12L:12D, 2L:22D, 22L:2D). Melatonin secretion is highly sensitive to darkness, and an association was found between the length of the melatonin production and the lighting programme. It may be supposed that changing the daily light and dark periods twice also affected melatonin production. Disturbing the activity of the pineal gland with the use of the 8L:4D:8L:4D photoperiod may have an effect on nursing behaviour.

Applying the intermittent lighting however strongly disturbed the nursing behaviour of the does and the nursing event occurred in every part of the day (dark and light periods). Irregular occurrence of nursing throughout the day can work against the animals' well-being, as according to Hudson and Distel (1982) the nursing times (of the doe) and suckling (of the kits) are harmonised and the suckling kits are waiting for their dam. Matics *et al.* (2004) observed that the nursing time of the does which nurse the kits twice a day may occasionally be longer than that of the does nursing once a day. It may be assumed that in this case the suckling kits were not expecting the does' arrival and did not leave the nest (they did not move to the top of the hair in the nest) as described by Hudson and Distel (1982). It was probably due to similar reasons that in the 8+8L group, where the nursing could happen during any part of the day, longer nursing times were recorded. Thus, the does often entered the nest while their kits were sleeping in the nest. Waking up and leaving the nest material lengthened the nursing period (the time does spend in the nest box).

Effect of nursing method

Changing free nursing to controlled nursing 3 d prior to AI increased the kindling rate by 8.1 points of percentage and the number of kits born alive also increased. However, the suckling mortality also increased; no difference was therefore found for litter size between the 2 groups at 21 d. The body weight of the FS does was higher, which suggests a better condition.

The kindling rate was improved in almost every previous experiment. The magnitude of the difference between the control and biostimulated groups depended on the fertility rate of the control group. The improvement was 26, 15.3 and 12 points of percentage according to Eiben *et al.* (2004a) (compared to 33.3%), Bonanno *et al.* (2004) (compared to 44.1%) and Eiben *et al.* (2004b) (compared to 46%), respectively. In the present study, an 8.1 points of percentage increase was observed compared to the 72.5% kindling rate of the control group. It should be noted that Eiben *et al.* (2007) and Matics *et al.* (2004) found no significant differences, as the control does also showed good kindling rates (71.1 and 77.8%, respectively). The efficiency of changing the nursing method depends on the fertility rate without treatment. This relationship was also examined from another angle. Applying PMSG treatment (Maertens, 1998), it was found that the kindling rate of the rabbit does significantly improved only after the 1st kindling when the energy balance of the does was negative and their feed intake was also limited (Xiccato, 1996). In the experiment of Bonanno *et al.* (2004), the improvement in kindling rate for the effect of a 2-d controlled nursing occurred in does with parity 1-3. In contrast, after the latter kindlings when the condition of the does is better the biostimulation may be inefficient.

When inseminating the does 11 d after parturition, due to the partial antagonism between the hormonal background of lactation and reproduction, the lactating does' performance was low (Theau-Clément, 2007). Ubilla *et al.* (2000) found lower plasma prolactin concentration 24 h after the doe-litter separation. According to Bonanno *et al.* (2004), the prolactin level can decrease after 20 h stimulating oestradiol-17 β secretion, which can induce oestrus. This may be the physiological background of the kindling rate increase caused by the change in nursing method.

In our experiment the body weight of the control group does was significantly higher than that of the experimental group (+0.14 kg). Similarly, slightly higher body weights were found in the control group by Eiben *et al.* (2004b, 2007). It may be assumed that because of the lower pregnancy rate of the control group the does that failed to conceive (due to low body condition) had a longer period for resting and so were able to regain their weight loss.

In line with our results in most experiments (Matics *et al.*, 2004; Eiben *et al.*, 2004ab, 2007) the total number of kits born and the number of kits born alive were higher in the biostimulated group, although the differences were not significant in most cases. Substantial improvement in litter size was recorded by Matics *et al.* (2004) when the controlled nursing followed the free nursing for 3 d (rather than for 2 d) prior to AI. Similarly to our results, no significant differences were found for litter size at 21 d or at weaning by Eiben *et al.* (2004a).

From the profitability viewpoint, the most important traits: the total number of kits born, total number of kits born alive, number of kits at 21 d and total kits' weight at 21 d per AI of the FS-CS group were higher than in the FS group by 16.2, 18.4, 9.3 and 6.3%, respectively. This finding shows that the biostimulated rabbits showed superior performances for all important traits. Compared to the control group, the difference found by this study was lower than that of Eiben *et al.* (2004a,b), but in those studies the kindling rates of the control groups were very low (33.3 vs. 46.0%) and greater increases could be achieved than in this experiment, where the control group kindling rate was 72.5%.

Compared to dam-litter separation (Theau-Clément, 2007), changing the nursing method was more favourable (Matics *et al.*, 2004; Bonanno *et al.*, 2004; Eiben *et al.*, 2004b) because in this case the body weight of the suckling kits did not decrease. However, similarly to our study, Eiben *et al.* (2004b, 2007) also found significantly lower body weight at 21 d in the biostimulated group. Presumably the suckling kits wanted to suckle at the accustomed time even after the change of the nursing method, generally during the hours after midnight. As observed by Hudson and Distel (1982), when the nursing events were not realised (the does were not allowed to enter the nest boxes) the suckling kits shortly re-entered their nests and slept calmly. When the does were allowed to enter the nest boxes in the morning, they probably found kits unprepared for nursing and covered with nest material. At these times, probably not all kits can reach the teat in time and suckle, so they may show some delay in body weight. The 21-d weight of FS-CS litters may also be lowered because controlled nursing avoids the multiple nursing events occurring, so on the whole kits suckle less milk.

It was previously reported by Matics *et al.* (2004) that the frequencies of nursing twice-a-day and 3 times a day were increased if the controlled nursing was changed to free nursing after 9 d and at the same time more does nursed their kits during early morning or during the morning hours. Our results show that when controlled nursing was practised for 3 d prior to AI at day 11, the frequency of the multiple daily nursing events increased after day 11 (between days 11 and 17)

and a second nursing peak appeared between 8:00 and 12:00 h. Those does that nurse their kits twice a day generally enter the nest boxes at nightfall, then during the morning hours.

The duration of the nursing event was longer for the biostimulated does than that of the FS does independently of the number of daily nursing events (first or second event) or of changing of nursing methods in group FS-CS. Those does that nursed their kits twice a day stayed in the nest boxes longer compared to those with a single nursing event per day. This phenomenon is caused by the suckling kits covered with hair not prepared for the arrival of their dams (Matics *et al.*, 2004). There is no clear explanation for the longer nursing events of the FS-CS does compared to the FS group recorded prior to the changing of the nursing method (when both groups could freely nurse the kits). It may be assumed that at the preceding kindling, changing the nursing method resulted in longer nursing events (especially for the does nursing their kits twice a day) which could affect the nursing behaviour after the following parturition. This hypothesis however requires further analysis.

CONCLUSION

The intermittent 8L:4D:8L:4D lighting programme is not advantageous from the production viewpoint and disturbs the nursing behaviour of does and suckling behaviour of kits (irregular occurrence of nursing throughout the day), which is disadvantageous for the well-being of the rabbits, so its application cannot be advocated.

Our experiment showed that changing the nursing method (biostimulation) can increase the kindling rate and the production per AI. Contrary to the results of previous experiments, the body weight of the suckling kits slightly decreased. Nevertheless, changing the free nursing to controlled nursing 3 d prior to AI can also be recommended in practice as a biostimulation method, since it allows a +6.3% gain in productivity per insemination.

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