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EFFECT OF HYPOKINESIA ON THE BEHAVIOUR OF LAMBS

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Summary

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The studies were performed on 13 lambs at the age of 3.5 months divided into 2 groups: experimental (n=6), immobilized for 72 h and control (n=7) that was allowed to move freely. The following ethological parameters: duration and dynamics of feeding, times spent lying and ruminating; times spent standing and ruminating, times spent lying or standing were evaluated by individual chronometry. The haematological parameters (red blood cell counts, white blood cell counts, haemoglobin content and haematocrit) were measured on an automated analyser System 150. Blood samples were obtained prior to immobilization (background) and by post immobilization hours 0.25, 24, 48 and 72.

Haematological data showed no statistically significant differences between groups during the entire experimental period. The most pronounced differences were observed by respect to the time spent standing and ruminating (a marker of discomfort) whose values were considerably higher in immobilized lambs than in controls. In experimental animals, a significant reduction in the time spent lying and prolongation of the time spent standing were also observed.

Key words: behaviour, haematological parameters, hypokinesia, lambs

INTRODUCTION

The possibilities for adaptation of animals to various environmental factors and their behavioural reactions are very important from both economical and medical point of view (Kvetnansky *et al.*, 1979; Nedava *et al.*, 1985; Iliev *et al.*, 1995).

The unfavourable impact of immobilization is manifested not only by restriction of motor activity, because the impaired individual distance is also responsible for the permanent tension of animals from the low hierarchical positions (Carter *et al.*, 1983; Grandin, 1983; Grandin *et al.*, 1986). An altered social behaviour is also observed in sheep from various breeds placed in a limited space due to their different requirements for individual and social distance, resulting in an altered feeding behaviour as well (Tosev *et al.*, 1989; Lynch *et al.*, 1992a).

The limitation of space is considered to be a physiological stressor in sheep (Lynch *et al.*, 1992b). Hutson (1981, 1984) reported that sheep could be extremely tolerant ranging paralelly one by another when the space is insufficient.

The contemporary sheep breeding necessitates the housing of sheep in a limited space thus placing the animals into new interrelationships both among them and with the environment, some of them being incompatible with ovine welfare (Fitko *et al.*, 1988; Hargreaves and Hutson, 1990; Iakubowski *et al.*, 1993; Minton *et al.*, 1995). At the same time, little is known about the mechanisms involved in the adaptation of ruminants to restricted motor activity. Therefore, specific studies on different ethological parameters in lambs, submitted to restriction stress are still needed.

The aim of the present study was to investigate the influence of hypokinesia on the behaviour of lambs and to test the hypothesis that the marked restriction of motor activity affected some ethological parameters in small ruminants.

MATERIALS AND METHODS

Experimental animals

The experiments were performed in June and July with 13 male lambs from the Stara Zagora breed at the age of 3.5 months. Prior to the experiment, the animals were placed in a 20 m^2 premise with a walking area of 30 m^2. The lambs were fed with 300 g concentrated forage given at 8:00 AM, alfalfa hay and water *ad libitum*. An accomodation period of 7 days was allowed.

Experimental design

The animals were divided into 2 groups – experimental (immobilized) – 6 animals with a total body weight of 125.3 kg and control one (non-immobilized) – 7 animals with total body weight of 111.2 kg. Both groups were housed in the same premises that were also used for accomodation prior to the experiment. The immobilization of experimental lambs was performed in a 150/80 cm cage i.e. a 0.2 m^2 individual area was assured. The feeding and drinking devices were mounted on one of the bigger walls of the cage. Thus, the animals were close one to another without possibility to move freely, and were allowed only to turn with difficulty, to eat and to drink. The immobilization lasted 72 h.

Lambs from both groups had a visual and auditory contact among them and were fed the same diet as during the accomodation period.

Ethological studies

The behaviour of lambs was monitored by individual, continuous chronometry for 96 h: 24 h prior to the immobilization (background) and 72 h afterwards (experimental period).

The feeding and resting behaviour were monitored via their components – foraging, rumination, static active state (standing) and rest (lying). The behavioural stereotypes: manifestations of discomfort. abnormal behaviour, communication and postures for maintenance of hierarchy order were noted.

The days were conditionally divided into two 12-hour zones – light and dark, called day and night respectively.

Haematological studies

Blood samples of 3 mL were obtained from *v. jugularis* during the morning feeding 24 h prior to (background, hour 0) and during the experimental period – hours 0.25, 48 and 72, in heparinized tubes. The following haematological parameters – red blood cell counts (RBC), white blood cell counts (WBC), haemoglobin content and haematocrit, were measured on an automated analyser System 150.

Statistical analysis

Comparisons of data within and between groups were performed by the least significance difference (LSD) t–test at a level of significance $p<0.05$.

RESULTS AND DISCUSSION

Feeding behaviour

According to generally accepted classifications, the feeding behaviour includes two elements, used as markers of ruminant welfare: feeding and rumination, the latter being especially important.

Feeding. The data from Table 1 showed that during the background period the feeding time in control and experimental sheep comprised about 35.9% to 39.75% of the day, respectively. Those values are higher compared to reference ones reported by others (Hofmann and Schwartz, 1995; Lankin, 1997; Baumont *et al.*, 1997; van der Heide *et al.*, 1999). This could be explained by the occurring changes in feeding during accomodation vs the period prior to the accomodation. During the accomodation, including background, the animals were fed *ad libitum* and this could be the cause for the increased feeding activity. It was the highest in hours immediately after the placement of forage but was high during nighttime as well. The difference between both groups during the background evaluation was statistically insignificant.

During the experimental period, there was a tendency towards decrease in feeding activity in both groups, especially in the immobilized one – by about 8% (Table 1). This change could be attributed to a certain suppression of hunger centre after the ingestion of a sufficient amount of nutrients. There was no significant influence of immobilization on feeding ac-

Table 1. Principal ethological parameters (mean±SEM) in non-immobilized (control group, n=7) and immobilized lambs (experimental group, n=7)

Behavioural activity[1]	Background[2]		Experimental period[3]	
	Control group	Immobilized group	Control group	Immobilized group
Feeding time	517.40 ± 19.19	572.40 ± 20.80	413.05 ± 17.14	434.97 ± 16.45
Time spent lying and ruminating	481.80 ± 16.16	418.40 ± 15.24	382.83 ± 11.26	303.27 ± 10.85
Time spent standing and ruminating	28.60 ± 2.54	20.80 ± 1.91	20.33 ± 1.28	135.23 ± 3.95 [b] [**]
Time spent lying	295.50 ± 9.64	300.50 ± 9.96	414.97 ± 12.95	303.37 ± 9.03
Time spent standing	115.17 ± 3.95	122.80 ± 5.62	107.30 ± 4.99	229.23 ± 7.76 [a] [*]

[1] All ethological parameters are given as minutes per animal per 24 hours; [2] Values obtained during the 24-hour period prior to the experimental period; [3] Values obtained during the 72-hour experimental period. Level of significance: [a] $p<0.05$; [b] $p<0.001$ between groups; [*] $p<0.05$; [**] $p<0.001$ vs background values.

Table 2. Red blood cell and white blood cell counts (mean±SEM) in non-immobilized (control group, n=7) and immobilized lambs (experimental group, n=7)

Time post immobilization, hours	Red blood cells, T/L		White blood cells, G/L	
	Control group	Immobilized group	Control group	Immobilized group
0	6.7±0.63 ^{a}	7.6±0.56 ^{f}	7.68±1.12	8.28±1.6
0.25	7.4±0.22 ^{c}	6.4±0.37 ^{e}	7.47±0.74	7.89±0.69
24	8.4±0.91	9.7±0.50 ^{d}	8.86±0.75	9.32±0.82
48	8.7±0.45 ^{b}	7.4±0.63 ^{e}	8.10±0.90	9.90±0.92
72	6.7±0.91	7.6±1.18	6.90±1.32	7.53±1.75

Level of significance within groups: $^{a\text{-}b;\ b\text{-}c;\ d\text{-}f}$ $p < 0.05$; $^{d\text{-}e}$ $p < 0.01$.

tivity, in contrast to the studies of Lynch *et al.* (1992b) where sheep were shown to compensate environmental stressors through increased feeding activity. Probably, the feeding activity in sheep was dependent on the age as well as on the nature, duration and intensity of the stressor.

Rumination. A primary parameter for discomfort evaluation was the position of sheep during rumination. It is known that when the time spent ruminating and standing is more than 40% out of the total rumination time, there is a factor acting negatively on animal physiology.

Normally, rumination occurs *in lying position* and during nighttime (Lynch, 1992a). Our findings during the background period were in conformity with these data (Table 1). In both studied groups, rumination time in this period was higher than during the experimental one. This is understandable in view of the increased feeding activity. The lower time of rumination in lying position during the experimental period was better manifested in immobilized animals.

There were also periods with increased rumination activity several hours after the feeding and especially during the night hours (from 9:00 PM until 8:00 AM).

The time spent for rumination *in standing position* by immobilized lambs was increased several times. Simultaneously, some changes during this period were observed. After the first hours following the immobilization, experimental lambs became accustomed to the conditions and achieved a synchronization so that most of them (4–5 on the average) succeeded to lie down on the limited space. The data for the 72-hour period (Table 1) showed that the duration of this parameter on experimental lambs was 6 times higher vs the background and nearly 5 times higher vs controls. The differences were with a high statistical significance ($p<0.001$) both vs the control group and vs background. At the end of the experiment, all animals succeeded to lie down, two of them using the feeding device for the purpose. This confirmed the hypothesis that the time for rumination in standing position was the most reliable marker of discomfort in ruminants (Lynch *et al.*, 1992a; 1992b).

Rest

It is generally accepted that in such studies, the components of rest are times spent lying and standing. The summarized results for both activities showed similar values in both studied groups during the background and the experimental period (Table 1) – 410.67 min and 522.27 min in controls and 423.3 min and 526.9 min in immobilized lambs, respectively. A clear tendency towards increased time spent lying was observed in controls during the experimental period vs the background (almost by 80%) but the respective values in experimental animals were almost identical (Table 1). The probable cause for those data in control lambs was the complete accomodation to the environment. The changes in immobilized lambs were opposite – 80% increase in standing activity vs both background and control data (Table 1).

Haematological changes

There were no significant differences in RBC counts between groups (Table 2). In immobilized lambs, RBC counts increased considerably by post immobilization hour 24 compared to hour 0 (9.7 ± 0.5 T/L vs 7.6 ±0.56 T/L, $P<0.05$) as well as to post immobilization hours 0.25 (6.4±0.37 T/L, $p<0.01$) and 48 (7.4±0.63 T/L, $p<0.01$). In control lambs, RBC dynamics was less variable and the values ranged between 6.7±0.63 T/L at the beginning of the experiment to 8.7±0.45 T/L by post immobilization hour 48.

The average baseline WBC counts in controls and experimental lambs were 7.68±1.12 G/L and 8.28±1.67 G/L, respectively. Fifteen minutes after the immobilization, they decreased to 7.89±0.69 G/L whereas in controls they remained almost unchanged (7.49±0.74 G/L). By post immobilization hour 24, WBC increased in both groups, although not statistically significantly. This trend was preserved 48 hours after the immobilization. Afterwards, at post immobilization hour 72, WBC counts decreased under the baseline ones in both groups. The differences within and between groups during the entire experimental period were not significant.

Haemoglobin content in both groups varied in narrow limits: 93±3.1 g/L – 113 ± 3.4 g/L in the experimental one and 97±4.6 g/L – 113 ±5.3 g/L in control one. Haematocrit changes were more substantial in immobilized lambs – from 0.30±0.02 L/L prior to immobilization to 0.26±0.01 L/L by post immobilization hour 72.

The changes in haematological parameters suggested that the used model of hypokinesia did not result in significant physiological changes. At the same time, the intensity of chosen model was sufficient to produce a functional increase in adrenal activity, as indicated by the statistically significant changes in cortisol, catecholamines and blood glucose concentrations in immobilized animals, especially during the first 2 hours (Iliev *et al.*, 1998; Kutsarov *et al.*, unpublished data).

The increase in RBC and WBC counts in experimental lambs by post immobilization hour 24 could be attributed to the increased activity of the sympathetic nervous system that is known to play a primary role in the triggering of a cascade of adaptive reactions in response to various stressors (Dantzer and Mormede, 1979; 1983). In our case, the effect of the increased activity of this system on blood cellular elements in immobilized group was probably manifested rather by shifting of blood out of blood stores (spleen, liver, subcutis etc.) and its inclusion in systemic blood stream than by stimulation of leuko- and erythropoesis. This assumption is also confirmed by the fact that both WBC and RBC counts decreased in the subsequent periods of the study and by the lack of continuous changes in haematocrit values.

In conclusion, this study provides evidence that the immobilization of 3.5-month old lambs affected markedly the feeding and rest behaviour. The predominant changes were the increased time spent standing and ruminating, the time spent standing as well as the lower time spent lying. The 72-hour hypokinesia in 3.5-month-old lambs did not provoke significant changes in RBC, WBC counts, haemoglobin content and haematocrit values.

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