The influence of length day on melatonin and prolactin secretion during lactation in asesonal sheep

Abstract
The study demonstrated that during the period of milking, the duration of the daylight had a significant effect on the lactation parameters in ewes lambed in January and June. Marked differences were noted in the levels of milk production in sheep, during the period of shortening daylight, i.e. in autumn, as the sheep lambed in June produced 50% less milk than ewes lambed in January. The dwindling milk production at the time of a shortening photoperiod resulted from the different course of melatonin and prolactin secretion. In the period of shortening daylight, the concentration of melatonin increased, while the secretion of prolactin decreased, thereby contributing to a lower milk synthesis.

Key Words: melatonin, prolactin, lactation sheep

Introduction
The effect of light on the behaviour of animals has been common knowledge for a long time. In most of the animals studied to-date, the biosynthesis of melatonin was shown to follow the rhythm depending on changes in the light conditions. The highest concentrations of this hormone, often dubbed “the hormone of darkness” are found at night (REITER, 1991 a, b; ARENDT, 1986). The role of melatonin was mostly associated with its effects on the reproductive system. The most recent studies have proven, however, that melatonin can also modulate the concentrations of prolactin, which is one of the principal hormones responsible for triggering and maintaining lactation in mammals. The hormonal status of these animals (rams), so different from that of lactating females, cannot help in elucidating precisely the relationships that exist in a light-lactation-milk production system (MORGAN et al., 1994; LINCOLN et al., 1995; MISZTAL et al., 1999). This study has therefore been designed to discover the effect of changing the lambing date (changing of day length) on the secretion of melatonin and prolactin, in connection with the milk production levels in an aseasonal sheep race, i.e. the Polish Merino.

Material and Methods
The studies were conducted in the Experimental Station of the Department of Sheep and Goat Breeding, at the Agricultural University in Krakow. The experiments involved 40 ewes from the Polish Merino race, aged 4-5 years and weighing 60 ± 5 kg. The ewes were randomly allocated to two groups, with 20 animals per group. Mating after oestrus synchronization was performed on 5th September for the first group (Group I) of sheep and on 15th January for the second (Group II). In both cases, the Polish Merino rams weighing 85±5 kg were used. The ewes of Group I lambed from 20th-25th January while of Group II – from 18th-24th June. From the pre-mating preparation period up to the 4th month of the pregnancy, ewes were fed according to standards, with traditional seasonal fodder (forage pasture, hay and an additional feed
concentrate (NORMS, 1993). From the 5th month of pregnancy to drying off, sheep received a 1.5 kg pelleted diet (7.5 MJ-net energy and 220 g crude protein) and an ad libitum hay supplement. Throughout the experiments, these sheep as well as those in the first experiment were kept indoors but allowed to use outside runs during the day. Lambs stayed with their mothers until 56 days of age, after which they were weaned and ewes were used for milking. When the offspring were reared, the milk production of sheep was estimated based on the weight gains of the lambs from 2 to 28 days of age, using a conversion factor of 4.5 l milk per kg weight gain of lamb. During the period of milk use, ewes were milked twice a day using an Alfa-Laval milking machine. Milk yields were recorded individually at 10-day intervals. From day 20 of lactation to drying off, blood was drawn from the sheep every 30 day to monitor the changes in prolactin and melatonin secretion. Blood samples were collected from the time of sunset over a period of 6 h, at 60 min. intervals, through a catheter inserted into the jugular vein 6 h before. After centrifugation in heparinized tubes, plasma was stored at -20°C until hormone assays were performed.

Analytical Techniques
Concentrations of prolactin and melatonin in the blood plasma were measured by radioimmunoassay techniques. The plasma prolactin concentration was assayed by the radioimmunoassay (RIA) double-antibody method, using antiovine-prolactin and antirabbit-gammaglobulin antisera according to WOLINSKA et al. (1977). Melatonin was assayed in unextracted plasma according to the method of FRASER et al. (1983), modified by MISZTAL et al. (1996). The milk performance parameters and plasma prolactin and melatonin concentrations are expressed as means ± SEM. The Mann-Whitney test was used to assay the differences between groups in the parameters of lactations. The effects of the treatments on the hormone concentrations were analyzed by one-way analysis of variation (ANOVA). An a priori level of statistical significance was set at P<0.05 for all tests. Post-hoc differences were determined using Scheffe’s test.

Results
The studies showed the significant effect of the lambing date on the lactation parameters in ewes during the milking period. The sheep lambed in January (Group I) were milked for an average of 104 days, whereas those lambed in June (Group II) – only for 66 days. In these milking periods, the Group I ewes produced 37.5 ± 9.6 l of milk, whereas those lambed in June – less than 50%, i.e. a mere 15.8 ± 5.1 l. (Fig. 1). The profile of prolactin secretion in the Merino sheep lambed in January showed a higher diversity compared with the changes in melatonin concentrations (Fig. 2). With the lengthening photoperiod, the melatonin level lowered and in May it amounted to a mere 61.0 ± 60.5 pg/ml, whereas in the same period, the prolactin concentrations was at its highest, reaching 298.7 ± 74.0 ng/ml. In July, yet another peak in prolactin levels (217.4 ± 68.3 ng/ml) was noted, again statistically significantly different (P ≤ 0.05) from other collections, except for the sample collected in June (180.3 ± 69.2 ng/ml). The melatonin concentrations did not show significant differences compared with the previous value (67.3 ± 40.7 pg/ml and 61.0 ± 60.5 pg/ml). However, from July till September, the melatonin secretion was more intense. In September, the concentration
Fig. 1: Mean (± SEM) monthly milk yield of Merino sheep lambed in January (group 1), and June, (group 2). See text for statistical comparisons.

Fig. 2: Mean (± SEM) plasma melatonin and prolactin concentrations in Merino sheep lambed in January. See text for statistical comparisons.

Fig. 3: Mean (± SEM) plasma melatonin and prolactin concentrations in Merino sheep lambed in June. See text for statistical comparisons.

of this hormone amounted to 85.3 ± 56.0 pg/ml, whereas the prolactin level decreased to 77.1 ± 57.5 pg/ml, so perhaps also contributing to the decrease in milk production (Fig. 2). The highest daily milk yield in ewes lambed in January was observed in May.
where it was 0.49 ± 0.09 l/day on average, while the melatonin was at its lowest value of 61.0 ± 60.5 pg/ml. From June through to September, a decrease in the milk yield occurred from 0.37 ± 0.05 l/day to 0.01 l/day. The concentrations of melatonin over that period showed an upward tendency, from 71.2 ± 50.5 pg/ml in June to 85.3 ± 56.0 pg/ml in September. Over the period of milking of ewes, the melatonin concentrations were stabilised, with an upward tendency at the time of the shortening photoperiod.

The results of the studies, concerning the changes in melatonin and prolactin concentrations in Merino sheep lambed in June, indicated that the highest prolactin concentrations (251.8 ± 62.6 ng/ml) was found in the first sample collection which took place in July, with the statistically significant difference (P ≤ 0.05) compared with any other sample. The melatonin levels at that time (48.1 ± 38.9 pg/ml) were the lowest (Fig. 3). From July onwards, a systematic decrease in the concentrations of prolactin was observed, with a simultaneous increase in melatonin concentrations. In September, the melatonin level was 73.2 ± 45.0 pg/ml, whereas the prolactin level dropped evidently to a mere 85.09 ± 67.0 ng/ml, which was significantly less (P ≤ 0.05), compared with the first sample collected in July (251.8 ± 62.6 ng/ml) and the second sample collected in August (165.5 ± 87.4 ng/ml). At the time of the changing photoperiod to the increased duration of the dark phase, the prolactin concentrations decreased: in October it was 62.8 ± 42.5, ng/ml and again the difference was statistically significant (P ≤ 0.05), compared with the levels in samples collected in July (251.8 ± 62.6 ng/ml) and August (165.5 ng/ml ± 87.4); in this period the concentration of melatonin increased by 3.8 pg/ml compared with the previous concentrations and amounted to 73.8 ± 34.1 pg/ml. The lowest level of prolactin (22.3 ± 16.4 ng/ml) was noted in November, and this value was significantly different (P ≤ 0.05) from the concentrations found in July (251.8 ± 62.6 ng/ml), August (165.5 ± 87.4 ng/ml) and September (85.09 ± 67.0 ng/ml). The melatonin concentrations in the last collection in November did not differ significantly from that found in October, and was 73.2 ± 45.0 pg/ml. The profile of the melatonin concentrations was stabilized and the differences were statistically insignificant. The highest milk yield in this group of sheep (0.18 ± 0.07 l/day) was found in August, when the prolactin concentrations was 165.5 ± 87.4 ng/ml, and the melatonin concentrations was 72.0 ± 37.3 pg/ml. In the following month, the secretion of melatonin rose to 73.2 ± 45.0 pg/ml and the milk yield dropped to 0.14 ± 0.05 l/day. In October, the melatonin secretion was the most intensive (73.8 ± 34.1 pg/ml) while the daily milk yield dropped to 0.03 ± 0.06 l/day.

Discussion

The differences in lambing dates and the associated lengths of the photoperiod, affected significantly the lactation parameters in sheep during the time of their use for milk production. The sheep which began to lactate in January, could be milked for a longer time compared with ewes lambed in June, that produced 50% less milk compared with ewes lambed in January. The duration of lactation and milk productivity found in the presented study in ewes lambed in January, is comparable with the results obtained by OSIKOWSKI et al. (1999) and WAŻNA et al. (2000). Ewes lambed in January, demonstrated the highest milk productivity in their first months of milking i.e. in May; the concentration of prolactin was also highest in these months whereas the melatonin secretion was at the lowest level. In subsequent months the melatonin levels rose while that of prolactin declined, and the milk production
dropped as well. The ewes which started lactation in June, in their first month of milking, i.e. in August, showed a higher level of prolactin and lower melatonin concentrations than the ewes lambed in January. These differences can be explained by the physiology of the lactation process, as for the sheep of Group I, it was the first month of milking, whilst for the sheep of Group II – the first month of milking. The profile of seasonal melatonin secretion in the ewes of Group I has shown characteristics of an annual rhythm described by MISZTAL et al. (1997), with a systematic increase in secretion levels from June onwards, whereas both prolactin concentration and milk production decreased over the same period. The levels of prolactin and of milk production in ewes lambed in June in the first two months of milking, can be compared to these two parameters in ewes of Group I, i.e. lambed in January, in the two last months of milking. These results indicate that the shortening daylight brought about an increase in melatonin concentration and a decrease in prolactin secretion, in sheep beginning lactation in January and June. The studies by GUT et al., (2001) confirmed the possibility of maintaining milk production in the Merino sheep throughout the year, but the lactation of sheep lambed in May and June lasted for only four months, while in those lambed in January it lasted for ca. ten months. Our own research corroborates these results, as the moving of the lambing date to the spring and summer months in aseasonal sheep exerted a conclusive impact on the lactation parameters during the milking period. Moreover, these results show that the lactation parameters are closely linked with the secretion of melatonin and prolactin. The literature data on the course of lactation in the Merino sheep lambed at various dates is scarce because the aseasonal characteristics of the race seemed to suggest, that the length of the photoperiod should not have a significant effect on milk production. Because of the absence of data on melatonin and prolactin profiles in lactating sheep, the results of this study can be related only to observations made in barren sheep (LINCOLN et al., 1995; MISZTAL et al., 1997). There is an interesting observation of the fact that the melatonin secretion in the aseasonal sheep studied, maintained similar levels from February till October, and that despite this, there was a drop in prolactin concentration and an inhibition of milk synthesis during the period of shortening daylight. This type of relationship can only be explained by the increasing role played by the biochemical signal from melatonin, prolonged in the period of shortening daylight and therefore resulting in decreased secretion of prolactin in response to it (REITER, 1991a; MORGAN et al., 1992; TRICOIRE et al., 2002).

References


Authors’ addresses
Dr. EDYTA MOLIK Prof. Dr. habil. EDWARD WIERZCHOŚ
Department of Sheep and Goat Breeding,
Agricultural University in Krakow,
Mickiewicza 24/28 Street, 30-059 KRAKOW, POLAND

E-Mail: rzmolik@cyf.kr.edu.pl

Dr. habil. TOMASZ MISZTAL
Dr. KATARZYNA ROMANOWICZ
The Kielanowski Institute of Animal Physiology and Nutrition
Polish Academy of Sciences
05-110 Jabłonna n/WARSZAW, POLAND