

Prolactin and Growth Hormone Circadian Periodicity in Lactating Cows¹ (36601)

J. A. KOPROWSKI,² H. A. TUCKER, AND E. M. CONVEY

*Animal Reproduction Laboratory, Department of Dairy Science, Michigan State University,
East Lansing, Michigan 48823*

Variations in ambient temperature and length of light markedly influence reproductive activity of many species of mammals (1, 2). For example, ovulation in rats and mice is linked closely to circadian changes in light and dark (3). In addition, luteinizing hormone in hens (4), rats (5) and possibly ewes (6) varies according to a circadian pattern. Direct evidence for changing serum concentrations of prolactin according to circadian rhythms has been reported in lactating (7) and adult male (5) rats and possibly in heifers during the estrous cycle (8). Serum growth hormone increases in humans during the late afternoon (9, 10), but there is no circadian periodicity associated with serum growth hormone in rats (11). The objective of this research was to determine circadian periodicity in serum prolactin and growth hormone in lactating cows.

Materials and Methods. Polyethylene (Intramedic-PE 190, Clay Adams, Inc.) or polyvinyl IV (Clay Adams, Inc.) cannulas were inserted into a jugular vein of cows at least 24 hr before the start of the experiment. To minimize hormonal fluctuations that may result from superfluous external stimuli throughout the study, the barn was restricted to the experimenter and to personnel that fed or milked the cows. Blood samples collected within 30 min after milking were discarded, because this stimulus is known to release prolactin and about 30 min is required for basal concentrations to be reestablished (12). The cows were fed at their usual times but if the feeding conflicted with a scheduled bleeding it was initiated after the

blood samples were collected. In the first experiment, blood was collected from 6 first-lactation Holstein cows at 0.5-hr intervals for 25 consecutive hr beginning at 7:00 a.m. In a second experiment, blood was collected at hourly intervals from four multiparous lactating Holstein cows for 72 hr. Blood samples were allowed to clot at room temperature for 2–4 hr, stored for 15–20 hr at 5°, centrifuged at 7000g for 10 min, and the serum was subsequently stored at –20°. Prolactin and GH were determined by radioimmunoassays of dilution duplicates of each serum sample as previously reported (12–14). Bovine GH (NIH-B₁₂) and prolactin (NIH-B₁) were used as reference standards.³

Inspection of the results of the two experiments revealed that the pattern of the changes in prolactin and growth hormone concentrations throughout the day were similar in both experiments. Thus, the data from the two experiments for each hormone were combined. The 0.5-hr values in the first experiment were pooled with the previous hourly sample (*i.e.*, 7:00 and 7:30 a.m. values were averaged and plotted as a 7:00 a.m. value). The combined data were analyzed by the least-squares multiple regression method.

Results and Discussion. Prolactin averaged about 28 ng/ml between 7 and 10 a.m., increased to a peak of 58 ng/ml at 4 p.m., declined irregularly to 28 ng/ml at 4 a.m. and then stabilized at about 28 ng/ml until 10 a.m. (Fig. 1). These data fit a quadratic regression equation ($p < .01$), but circadian patterns account for only 8% of the total variation in serum prolactin.

The circadian pattern of rising serum pro-

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lactin during the middle to late part of the light period of the day agrees with similar data in lactating rats (7). Schams and Karg (15) also observed marked changes in the concentration of serum prolactin within cows, but did not ascertain if circadian patterns contributed to the variation. Swanson and Hafs (8) reported that serum prolactin was highest in virgin heifers during the light hours, with the exception of a marked decrease at noon, and lowest during the night; they commented that their diurnal data may have been biased because the samples were collected in heifers only around estrus.

Inspection of the data from each cow strongly suggests that prolactin is released in pulses throughout the day. Serum prolactin in a typical cow is shown in Fig. 2. Partitioning the variance from the least-squares analysis revealed that differences within-cows accounted for 45% of the total variation whereas differences among-cows accounted for 47%. Since within-cow and circadian variance account for 53 % ($45 + 8$) of the total variation, both variables should be recognized in the design of future studies of prolactin in the bovine.

Although serum growth hormone in individual cows varied as much as 300% between consecutive hourly samples in a few isolated

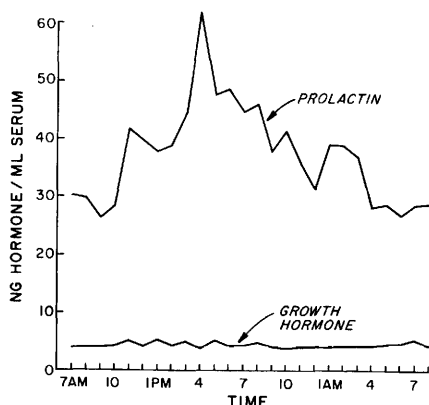


FIG. 1. Serum prolactin and growth hormone of lactating cows during a consecutive 25-hr sampling period. Prolactin data fit a quadratic curve ($p < .01$). Standard errors of mean ($n \cong 20$ at each point) for prolactin ranged from 1.3 to 7.9 and was proportional to the mean. Standard error of mean for growth hormone ranged from 0.3 to 0.9.

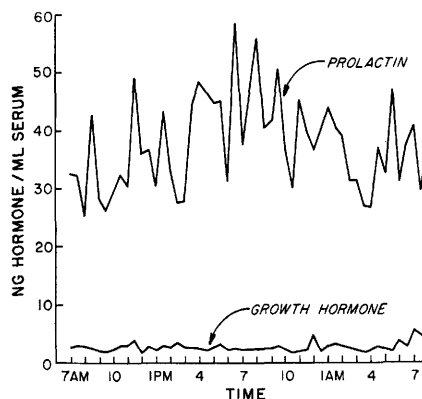


FIG. 2. Serum prolactin and growth hormone of an individual cow during a consecutive 25-hr sampling period.

instances, growth hormone generally was less variable than prolactin (Fig. 2). In contrast to prolactin, growth hormone did not vary according to a circadian pattern (Fig. 1). This agrees with data collected in rats (11), but is in contrast to the circadian periodicity reported in humans (9, 10).

Summary. Prolactin and growth hormone in serum of 10 lactating cows was measured by radioimmunoassay at hourly intervals on 4 days. Prolactin in lactating cows was released in erratic pulses throughout the day. Serum prolactin concentration followed a 24-hr periodicity ($p < .01$), with the highest value occurring at 4:00 p.m. (58 ng/ml) and lowest values being recorded between 4 and 10 a.m. (28 ng/ml). However, circadian changes accounted for only 8% of the total variation in prolactin values, differences within cows accounted for 45% and differences among cows accounted for 47%. Serum growth hormone concentration was more stable than prolactin and did not exhibit circadian periodicity.

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