ABSTRACT. Seasonal levels of LH, FSH, testosterone (T), estradiol, progesterone (P), and prolactin (PRL) were determined in the plasma of five adult bulls, and five barren and four pregnant cows of Alaskan reindeer (Rangifer tarandus), which were sampled every 3 weeks for 54 weeks. The male reproductive axis was sequentially activated; LH peaked in May–June (2 ng/ml), FSH in June (51 ng/ml), and T in September (11.8 ng/ml). LH levels in females reached a maximum in both groups at the end of August (the beginning of the rut). Seasonal variation in FSH was minimal in pregnant cows, but exhibited one elevation (41 ng/ml) in barren ones in November. T levels in cows remained at barely detectable levels. The decrease of T values observed in both groups in December and March was not significant. PRL peaked in May in cows (135 ng/ml pregnant, 140 ng/ml non-pregnant) and in June in bulls (92 ng/ml). Estradiol was highest in bulls in the rut (August), in non-pregnant cows in January and in pregnant cows in April, shortly before parturition. P levels in the pregnant cows rose from September and peaked (9 ng/ml) shortly before parturition in April. In the non-pregnant females P values increased and decreased several times before peaking (5 ng/ml) in March. In the males, the variation of T and estradiol levels correlated relatively well with the antler cycle but in the females the variation of neither estradiol, progesterone nor T appeared to be related to mineralization or casting of antlers. Copyright © 1997 Elsevier Science Inc. comp biochem physiol 116B;2:269–277, 1997.

KEY WORDS. LH, FSH, testosterone, prolactin, estradiol, progesterone, reindeer, pregnancy, seasonality, antler cycle

INTRODUCTION

Seasonal variation of reproductive hormones [LH, FSH, testosterone (T), and prolactin (PRL)] has been investigated frequently in various cervids of temperate regions [white-tailed deer (9,10,28); roe deer (42); red deer (2,18); Pere David’s deer (25); fallow deer (3)]. Conversely, studies of the reproductive hormones in the boreal cervids, e.g., moose, reindeer, and caribou are relatively rare (4,23,26,40,57).

Reindeer/caribou (Rangifer tarandus) is the most northern cervid and the only one in which the females carry antlers. Whereas the regulation of the antler cycle in male deer of temperate zones has been studied extensively (7,21,49), and is well understood, this is not the case for male and female reindeer (22,23,26,39–41,51,57,59).

In most cervids, the seasonal variation of T is the predominant factor in determination of individual phases of the antler cycle, such as the growth period and the time of polishing of the velvet and casting of antlers (9,19,24,42). In the male reindeer this correlation between T levels and the antler cycle is less pronounced. Although the growth of male reindeer antlers occurs also during the period of low concentrations of plasma T (58), out-of-phase polishing of antlers can be induced by administration of large doses of exogenous T (39) and antler casting will follow shortly after castration (50), the timing of the individual phases of the antler cycle does not entirely relate to the variation of T in plasma (57). In addition, unlike in most other cervids, antlers are produced in both sexes, the pedicles and antlers developed well before puberty and gonadectomy does not prevent the growth and subsequent replacement of antlers (23,50,59). In addition, long-term administration of an androgen receptor blocker cyproterone acetate which causes quick casting of antlers in other cervids (7,19), was not effective in the reindeer male or female calves or yearlings (G. Bubenik et al., unpublished observation). Therefore,
based on the above data, Lincoln and Tyler (23) concluded that in males “the secretion of T by testes, although not essential for the growth and seasonal replacement of antlers, has an overriding influence of the timing of the antler cycle.”

In the female reindeer the regulation of the antler cycle has been investigated only rarely (22,23). The results of these studies indicate that in intact females ovarian steroids, possibly estradiol, participate in antler mineralization. In ovariectomized animals the hardening of antlers may have been induced by androstenedione, which is most likely of adrenal origin (23).

In view of the many gaps which exist in the elucidation of the endocrine regulation of the antler cycle in the male and female reindeer we decided to perform a detailed investigation of the seasonal changes of reproductive hormones in relationship to antler cycles of both sexes. As the timing of casting of antlers differs in pregnant (PG) and non-pregnant (NP) cows (26), our present study aimed to investigate each group separately.

**MATERIAL AND METHODS**

**Animals and Sampling Procedure**

Experimental protocols for this study were approved by the University of Alaska Fairbanks campus-wide animal welfare committee (IACUC #92-016).

Fourteen adult reindeer (*Rangifer tarandus*) were housed outside at the Large Animal Research Station and at the Biological Reserve, Institute of Arctic Biology, Fairbanks, AK (latitude 66°N). The animals were divided into 3 groups: 2- to 6-year-old bulls (n = 5), 2- to 5-year-old non-pregnant cows (n = 4), and 2- to 5-year-old pregnant cows (n = 5). The females that were selected to become pregnant were kept with bulls during the entire rutting season and some time thereafter. The females selected to be non-pregnant were kept apart from bulls in a separate enclosure approximately 300 m away. All animals were kept in large paddocks with constant access to pasture grasses. They also received a diet composed of 85% textured pelleted reindeer feed (dry matter values ranged from 15–15.6% digestible protein, 9.6–10.8% acid detergent fiber, 77.5–78.5% TDN), and 15% chopped grass hay (dry matter values ranges from 5–9% digestible protein, 38.6–41% acid detergent fiber, 56.8–57.9% TDN). Consumption in reindeer varies seasonally (56) and ranged from approximately 1–4 kg/animal/day. Consumption of grass pasture was not measured. All animals were provided with ad libitum water in the summer and snow in the winter.

Every 3 weeks, from June 5, 1992 to June 18, 1993, each animal was immobilized using 1–2 mg/kg xylazine hydrochloride (Anased, Lloyd Labs. Shenandoah, IA, U.S.A.) and 1.5 mg/kg of ketamine hydrochloride (Ketaset, Aveco Co., Fort Dodge, IA, U.S.A.). Doses varied depending on individual animal temperament. Rutting bulls are exquisitely sensitive to xylazine and seem to be unable to properly clear ketamine (J. Blake, unpublished data), therefore they were immobilized using only 0.1–0.2 mg/kg of xylazine hydrochloride. Drugs were administered via hand injection, pole syringe, or blow pipe. The method of administration was determined by animal temperament and human safety consideration with the primary intent to minimize excitement. After blood collection and antler measurement the reindeer received an intravenous injection of yohimbine (0.125 mg/kg) to reverse the xylazine.

Once immobilized, three consecutive samples of blood (35 ml each) were drawn from the jugular vein into heparinized tubes at 10 min intervals. The blood was centrifuged within 4 hr of collection and plasma was separated and frozen at −20°C. Life history events were recorded for each animal throughout the study period. The antlers were usually polished within a two-day period and mostly at the same time. The casting was also mostly synchronous; in cases of a desychronous casting an average date of both drops was recorded. As the date of initiation of antler regrowth we have chosen the period when a complete re-epithelization of the pedicle wound occurred. This was followed almost immediately by a vigorous antler growth. The data of parturition were recorded daily for the PG group.

**Radioimmunoassays**

The concentrations of LH, FSH, testosterone, prolactin, estradiol, and progesterone were determined in plasma in duplicate by radioimmunoassays, according the techniques described in previous papers. For each hormone all samples were assayed at the same time.

LH was determined by a homologous bovine assay with no crossreactivity to other pituitary hormones (43,44). The reference preparation was a bovine pituitary LH-DSA. The sensitivity was 0.05 ng/tube; intra-assay coefficient of variation (CV) averaged 8.6%, and interassay variation was 11.5–14%. The reference preparation was a pure bovine pituitary extract prepared in our laboratory (LH-DSA with a biological activity of 1.0 times NIH-LH-S1). The assay was validated for reindeer plasma by recovery studies of 4 different concentrations added to plasma and was on average 97.5 ± 6.2%. Dilution curves of reindeer plasma with a high LH content ran parallel to bovine standard.

FSH was determined by a system where pure ovine FSH was used in labeling, and the antisera against ovine FSH was produced in guinea pigs (43,46). As a reference preparation we used USDA-bFSH-B1. The sensitivity was 5 ng/tube and intra-assay CV was 7.5%, an interassay CV was 12–18%. Prolactin was determined by a homologous bovine assay. The antisera produced in rabbits exhibited no crossreactivity to other pituitary hormones. The reference preparation used was USDA-bPRL-B1 (biological activity 0.49 ×
TABLE 1. Statistical values for analyzed hormones

<table>
<thead>
<tr>
<th>Hormone</th>
<th>Class</th>
<th>D.f.</th>
<th>F</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>LH</td>
<td>General model</td>
<td>61,823</td>
<td>17.98</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Category</td>
<td>2,823</td>
<td>90.26</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Date</td>
<td>54,823</td>
<td>16.29</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>3,823</td>
<td>1.98</td>
<td>NS</td>
</tr>
<tr>
<td>FSH</td>
<td>General model</td>
<td>61,823</td>
<td>18.51</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Category</td>
<td>2,823</td>
<td>171.23</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Date</td>
<td>54,823</td>
<td>13.94</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>3,823</td>
<td>13.91</td>
<td>0.001</td>
</tr>
<tr>
<td>Testost.</td>
<td>General model</td>
<td>61,823</td>
<td>12.17</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Category</td>
<td>2,823</td>
<td>60.74</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Date</td>
<td>54,823</td>
<td>11.04</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>3,823</td>
<td>2.14</td>
<td>0.09</td>
</tr>
<tr>
<td>Estradiol</td>
<td>General model</td>
<td>61,823</td>
<td>8.65</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Category</td>
<td>2,823</td>
<td>68.91</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Date</td>
<td>54,823</td>
<td>4.40</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>3,823</td>
<td>68.08</td>
<td>0.001</td>
</tr>
<tr>
<td>Progest.</td>
<td>General model</td>
<td>40,520</td>
<td>27.96</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Category</td>
<td>1,520</td>
<td>105.83</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Date</td>
<td>36,520</td>
<td>28.36</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>1,520</td>
<td>0.13</td>
<td>NS</td>
</tr>
<tr>
<td>Prolactin</td>
<td>General model</td>
<td>61,823</td>
<td>36.51</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Category</td>
<td>2,823</td>
<td>16.33</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Date</td>
<td>54,823</td>
<td>40.17</td>
<td>0.001</td>
</tr>
<tr>
<td></td>
<td>Age</td>
<td>3,823</td>
<td>4.41</td>
<td>0.01</td>
</tr>
</tbody>
</table>

There were overall differences for all hormones ($P < 0.001$). All hormones differed by Category ($P < 0.001$) and Date ($P < 0.001$). Age differences were found in some cases but trends were unclear and inconsistent (Table 1).

Residual correlation indicated significant relationship between gonadotropins and sexual hormones mostly in males. Fewer correlations were found for females (Table 2).

**Antler Cycles, Rut, and Parturition**

The range of dates of antler polishing, the beginning of antler growth, the casting, and the parturition are presented in Table 3.

**LH, T, and FSH in Bulls**

In bulls, LH reached peak levels (1.98 ng/ml) in May (late March vs April, $P < 0.001$, April vs May, $P < 0.001$) and the minimum (<0.1 ng/ml) was observed in January (late June vs July, $P < 0.001$, July vs early August $P < 0.001$). Mean T levels began to increase at the time when LH levels were already declining (Fig. 1). A steep increase of T followed in August, achieving peak levels (11.8–11.5 ng/ml) from the end of August until mid-September (late August
TABLE 2. Residual correlation coefficients between selected hormones by category

<table>
<thead>
<tr>
<th>Hormones:</th>
<th>LH × FSH</th>
<th>LH × PRL</th>
<th>FSH × PRL</th>
<th>LH × Test</th>
<th>Estr × LH</th>
<th>Estr × Test</th>
</tr>
</thead>
<tbody>
<tr>
<td>Males</td>
<td>0.39</td>
<td>0.19</td>
<td>0.20</td>
<td>0.14</td>
<td>0.10</td>
<td>0.38</td>
</tr>
<tr>
<td>(n = 303)</td>
<td>p &lt; 0.001</td>
<td>p &lt; 0.001</td>
<td>p &lt; 0.001</td>
<td>p &lt; 0.02</td>
<td>p &lt; 0.07</td>
<td>p &lt; 0.001</td>
</tr>
<tr>
<td>Females NP</td>
<td>0.13</td>
<td>0.03</td>
<td>0.25</td>
<td>0.18</td>
<td>−0.01</td>
<td>0.01</td>
</tr>
<tr>
<td>(n = 228)</td>
<td>p = 0.06</td>
<td>NS</td>
<td>p &lt; 0.001</td>
<td>p &lt; 0.01</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td>Females P</td>
<td>0.20</td>
<td>0.20</td>
<td>0.07</td>
<td>−0.05</td>
<td>0.01</td>
<td>−0.07</td>
</tr>
<tr>
<td>(n = 293)</td>
<td>p &lt; 0.001</td>
<td>p &lt; 0.001</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
</tbody>
</table>

vs early August (p < 0.001), the peak period of the rut. The FSH levels began to rise significantly in April and achieved maximum (51 ng/ml) in late May (April vs early May, p < 0.001, early May vs late May, p < 0.001) and June (Fig. 1). In males, T levels correlated relatively well with the development of antlers. The polishing occurred at the time of rising T levels and the antlers were cast at the time of minimal T concentrations. Interestingly, the highest concentrations of LH, FSH, and T were observed in the two most aggressive bulls.

**LH, FSH, and T in Cows**

In pregnant cows peak LH concentrations (0.84 ng/ml) were detected at the end of August, the beginning of the rut, and then declined to levels below 0.1 ng/ml. In the NP cows LH reached also a peak in August but, due to a large (pre-ovulatory) peak (7.3 ng/ml) detected in one female, the LSMEANS reached 2.3 ng/ml. A second LH peak observed in that group in November matched a similar peak of FSH (both LH peaks being significantly higher than any other value p < 0.001) (Fig. 1). The pregnant group exhibited a FSH peak (25 ng/ml) in mid-November (Fig. 1). Similarly, FSH concentrations in the NP group were almost identical to FSH values in the PG group except for a sharp peak (40 ng/ml) detected in November (this FSH peak was significantly higher than any other value p < 0.001) (Fig. 1). Variations of T levels in females (<0.05–0.3 ng/ml) was not related to the antler cycle either in the PG or in the NP females. Interestingly, in both female groups T levels exhibited a very similar seasonal pattern with an unexplained sharp decrease in December and May (Fig. 1, insert).

**Estradiol**

Peak estradiol (E₂) levels in bulls (3.5 pg/ml) were reached during the rut in mid-August (p < 0.001) (Fig. 2).

In NP cows E₂ reached a maximum (2 pg/ml) in January and in the PG females the peak concentration (3 pg/ml) was detected in April, shortly before the parturition (p < 0.001). Unlike in males, E₂ levels in females varied independently from the levels of testosterone (Table 2).

The elevation and decline of E₂ in bulls correlated with the mineralization and casting period of antlers, but a similar relationship was not seen in the females.

**Progesterone**

In the PG females, progesterone (P) concentrations reached peak levels (8.8 ng/ml) at the end of March, about one
TABLE 3. The range of dates of antler polishing, antler casting, new antler growth, and parturition

<table>
<thead>
<tr>
<th></th>
<th>Antlers polished</th>
<th>Antlers cast</th>
<th>New antler growth</th>
<th>Parturition</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Range (average)</td>
<td>Range (average)</td>
<td>Range (average)</td>
<td>Range (average)</td>
</tr>
<tr>
<td>Females</td>
<td>Aug. 30–Sep. 15 (Sep. 9)</td>
<td>NP Mar. 26–Apr. 9 (Apr. 2)</td>
<td>Apr. 22–Apr. 28 (Apr. 25)</td>
<td>Apr. 4–May 9 (Apr. 23)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>PG Apr. 4–May 1 (Apr. 24)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

NP = non-pregnant; PG = pregnant.

month before parturition. After parturition P levels dropped to non-detectable levels (<0.2 ng/ml) (P < 0.001).

In NP females, P concentrations increased and decreased several times before reaching peak levels (4.8 ng/ml) in March (Fig. 2). However, statistically the early March peak did not differ from the February values.

Prolactin

Prolactin concentration exhibited a typical seasonal variation with significant peak levels detected in all cows in May and bulls in May and June (Fig. 2). Both female groups reached almost identical peak concentrations (135 NP vs 140 ng/ml PG) as compared to the significantly lower peak of bulls (92 ng/ml). One female that gave birth 3 weeks before the rest of the group exhibited an early elevation of prolactin. However, during the late lactation period the concentrations in both female groups were very similar.

DISCUSSION

In previous studies, the seasonal levels of reproductive hormones investigated in reindeer of both sexes followed a pattern established earlier in other cervids. However, there was often little information available about the relationship between the peripheral levels of sexual steroids and the individual phases of the reindeer antler cycle.

Males

LH, FSH, T. In bulls, the sequential activation of the reproductive system starts with an almost simultaneous elevation of LH and FSH in April. LH achieved peak levels in May, about one month before FSH (Fig. 1). Similar to values reported by Stokkan and co-workers (51), LH values decline from May on and were already below 1 ng/ml during the rut. As in other cervids (9,12,49,53), T exhibited peak levels in August and September (i.e., the rutting season), more than 3 months after the LH maximum was detected. LSMEANS peak levels of T in our study (11.8 ng/ml) were in the range similar to 10 ng/ml observed by Stokkan and co-workers (51). Highest T levels (60 ng/ml) reported by Whitehead and McEwan (57) were also comparable with the highest concentration observed in one of our bulls (41 ng/ml).

Generally, the highest individual concentrations of LH, FSH, and, subsequently, T were detected in the most aggressive bulls, exhibiting also the largest antlers. These observations are in accordance with our study in white-tailed deer, where highest T and LH levels were detected in the most aggressive bucks of prime breeding age (10). Similar correlation between T concentrations and the fighting rank was reported in reindeer by Stokkan and co-workers (51).

The increase of T levels observed during the rapidly declining concentrations of LH is mostly due to an altered sensitivity of the reproductive system to the stimulation by
In the early stages of the reproductive activation a small dose of GnRH induces a rapid elevation of LH but a small increase of T; in the later stages, the same dose GnRH will cause a massive increase of T but a small one of LH (11,29,52,53).

Unlike in other cervids, some males did not finish the polishing of antlers before T levels reached a seasonal maximum of 11.8 ng/ml (see Fig. 1). Conversely, the casting of antlers occurred in some bulls more than 6 weeks after a decline of T levels to a baseline (<0.5 ng/ml). Similar delay in antler casting after a decline of T to basal levels (lasting for 2–3 months) was observed also in caribou by Whitehead and McEwan (57). This differs from the observations in white-tailed deer, where T levels detected during the time of antler cleaning (around 1.5 ng/ml) are similar to levels detected during the time of antler casting (7). In addition, antler casting was not achieved with an intramuscular administration of 100 or 200 mg/animal/week of androgen receptor blocker cyproterone acetate given to yearling male and female reindeer (G. Bubenik et al., unpublished observation). These data indicate that androgen receptors in reindeer antlers are relatively slow to respond to rapid changes of circulating androgen levels.

The onset of new antler growth varied considerably within our bull group (February 5–March 20) but it usually started about 8 weeks after casting (December 7–January 20). As expected, the two more aggressive bulls cast the antlers earlier than the three tamer ones. Our casting dates were in variance with the data of Lincoln and Tyler (23) who reported the casting of antlers in their bulls already in November–December but the new antler growth began only 3–4 months later, in April. This difference could be either due to the more northern latitude of norvegian reindeer (69.2°N), different nutritional conditions or due to the genetic differences between groups. Persisting individual differences in the casting dates (up to 2 months apart) have been reported in white-tailed deer (6).

**ESTRADIOL.** Maximum male estradiol levels (3.5 ng/ml), reached during the peak of the rut, were surprisingly higher than peak levels observed in pregnant females (3.0 ng/ml) detected shortly before parturition in April (Fig. 2). Similarly, concentrations of estradiol in the feces of caribou bulls were reported to be comparable to levels of non-pregnant cows (27). A significant correlation found between T and E2 may indicate that, similar to stallions (36), reindeer testes are aromatizing considerable amount of T into estradiol. Whether estrogens are involved in the mineralization of reindeer antlers has not yet been investigated, however experimental evidence indicates that E2 is 10–50X more potent in the mineralization of antlers than T (14,30). In addition, growing antlers exhibit estradiol receptors (20) and an experimental blockade of E2 receptors interrupts the mineralization process in the compact antler bone mantle, resulting in severe reduction of its thickness (8). Lincoln and Tyler (23) concluded that E2 is the main regulator of the antler cycle in the female reindeer and adrenal androgen androstenedione may be the secondary steroid involved in antlerogenesis. As the relationship between the testosterone and antler cycle in the male reindeer is less pronounced than in other cervids, it could be speculated that in addition to T, estradiol could play a role of a secondary steroid involved in the male reindeer antlerogenesis.

**Females**

**LH, FSH, T.** In females, the evaluation of hormonal data in relationship to their reproductive activity is difficult, as the 3 week period between the samples is too long to detect rapid changes in the fluctuation of reproductive hormones. In all cows, peak levels of LH were detected during the rut (28 August); however, the presence of 2 peaks of LH in the NP cows may indicate that subsequent estrous occurred in that group. This assumption is further supported by the data on FSH where in the NP cows several oscillations were detected before peak levels were observed on November 6 (Fig. 1).

The much higher elevation of average LH levels observed in the non-pregnant group was largely due to a massive increase of LH (7.2 ng/ml) observed in one cow. Such a rapid (preovulatory) increase of LH which lasts only several hours (3,33) is difficult to detect and was probably missed in most of our other animals. Seasonal variation of testosterone levels in both groups of females was remarkably similar, with both groups exhibiting a non-significant trend with a decrease in December and May. The time course of T was not in any case related to the milestones of antlerogenesis, such as the polishing or casting of antlers (Fig. 1, insert).

**ESTRADIOL.** Seasonal levels of estradiol stayed below 1 pg/ml for most of the year. Only in the pregnant cows, LSMEANS of E2 rose to 3 pg/ml at the beginning of parturition, three weeks after peak concentration of progesterone were detected (Fig. 2). This slow elevation of E2 was in agreement with Baksi and Newbrey (4) who reported that E2 concentrations in pregnant reindeer cows in January were only 50% higher than values detected in June. Similar slow rise in E2 levels in *Rangifer* was also reported by Messier and co-workers (27) who noted that until day 150 of pregnancy no difference was detected between E2 of pregnant and non-pregnant cows. Conversely, the rapid elevation of E2 shortly before parturition is in agreement with data reported by Blom and co-workers (5) in reindeer cows, Plotka and co-workers (32) in white-tailed does, and by Kelly and co-workers (18) in red deer hinds. The variation in LSMEANS of E2 concentration in our study (1–3 pg/ml) was very similar to data of Lincoln and Tyler (23) who reported average E2 values varying seasonally between 2–6 pg/ml. However, probably because of species differences,
variations in laboratory techniques and because of an infrequent sampling intervals, our peak values of $E_2$ (3 pg/ml) were much lower than the ones reported by Plotka and co-workers (31) in the white-tailed deer (180 pg/ml) or by Kelly and co-workers (18) in the red deer (35 pg/ml).

The rapid decline of $E_2$ levels, which correlated with the time of parturition was usually followed by the casting of antlers (Fig. 2). Similar to reports given by McEwen and Whitehead (26) and Lincoln and Tyler (23), the NP cows cast antlers earlier than the pregnant ones (Table 3). Based on the results in ovariectomized cows, Lincoln and Tyler (23) concluded that ovarian $E_2$ is the major steroid regulating the antler cycle in the female reindeer. This appears to be true in some females, which cast their antler after parturition. However, in our non-pregnant females no decline of $E_2$ was observed before their casting (Fig. 1) and in the pregnant group two cows cast their antlers several days before parturition. As on few occasions, we have observed antler casting in the pregnant cows more than 2 weeks before parturition and McEwan and Whitehead (26) reported a casting period 2 months before the calving, it was concluded that a sudden drop in $E_2$ levels is probably not the only factor initiating the casting of antlers in the female reindeer.

**PROGESTERONE.** The seasonal variations of progesterone concentrations detected in our pregnant cows (Fig. 2) confirmed the expected time course with highest levels (9 ng/ml), observed shortly before parturition (Table 3) [for review, see (38)]. Similarly to some bovids, ovaries are the essential source of $P$ in the pregnant cervid female (34). Peak $P$ levels (9 ng/ml) detected in pregnant reindeer, were higher than 5 ng/ml reported in pregnant red deer (2), 4.6 ng/ml found in white-tailed deer (31) or 3.2 ng/ml observed in roe deer by Schams et al. (43). However, they were very similar to 10.9 pg/ml reported in reindeer by Blom and co-workers (5) or 8.4 pg/ml observed in roe deer by Sempere (48). Whereas in red deer $P$ concentrations were maintained high for most of the second and third trimester and calving occurred only after $P$ dropped precipitously (2,18), in our reindeer $P$ levels rose steadily until peak levels in March, some 3–4 weeks before parturition and then declined sharply. Similar steady rise in $P$ values during the pregnancy, followed by parturition and a subsequent rapid decline, was also reported by Blom and co-workers (5).

In contrast, the peak levels in the NP cows were not only smaller (6.0 ng/ml) but also exhibited an irregular variation (Fig. 2). The time course of $P$ levels in our NP females may be the result of seasonal polyestrus demonstrated by Plotka and co-workers (33) in white-tailed deer and recently by Ropstadt and co-workers (37) in reindeer. Similarly, during the first trimester, peak $P$ levels in NP white-tailed does were not different from concentrations recorded in the pregnant ones (31). Similar data were reported by Messier and co-workers (27) who investigated $P$ in feces of PG and NP caribou during the first 50 days of pregnancy. Finally, $P$ levels were also found elevated in NP cows in the study of McEwen and Whitehead (26).

It could be argued that a sharp drop in progesterone levels at the end of pregnancy may be related to antler casting. However, whereas there is plenty of evidence about the role of estradiol in the antler cycle (as mentioned above), there is little evidence that progesterone levels can decisively influence antler mineralization and retention. Whereas Jaczewski (15) reported partial or complete velvet shedding and retention of antlers in some castrated red deer stags treated with large doses of $P$, neither Waldo and Wislocki (54) in white-tailed deer nor Goss (13) in sika deer were able to induce velvet shedding in castrated bucks treated with $P$. Furthermore, McEwen and Whitehead (26) observed antler casting in captive pregnant reindeer from January until March (the period of high $P$) whereas in the wild females the casting occurred mostly after calving in June or July (the period of very low $P$). Therefore, it appears that $P$ does not play a crucial role in the antler cycle of the female reindeer.

**PROLACTIN IN MALES AND FEMALES.** Prolactin levels exhibited a typical seasonal curve with peak levels observed in May and June and trough concentrations detected from September until March (12). In both female groups, maximum concentration exceeded that of the bulls (Fig. 2). This is in agreement with PRL data on white-tailed deer and red deer, where peak female levels were higher than ones observed in males (18,47). Time courses of PRL levels in the individual cows indicate that the period of parturition/lactation coincides with the increase of PRL levels, however a continuation of lactation in ruminants does not apparently require peak PRL concentrations (17). This was confirmed in our study, as the sharp decline in PRL levels was observed in both the lactating as well as the non-lactating cows. However, this finding was in contrast to data of Adam and co-workers (1) who, later in the lactation period, reported higher PRL concentrations in the plasma of lactating hinds, as compared to the non-lactating ones.

Because in some cervids new antler growth coincides with PRL increase in the spring, Lincoln and Tyler (23) suggested that PRL may be involved in the reinitiation of antler growth. This hypothesis is not supported by our data as a new antler growth was observed in some reindeer bulls already in February, the time of minimal levels of PRL detected in our study (see Fig. 2). In addition, new antler growth in tropical rusa deer is initiated during the decline of PRL levels (53) and in the subtropical Eld’s deer, PRL levels rise when the antlers are hard (29).

In conclusion, our studies indicate that in reindeer, the individual milestones of their antler cycle are less related to the seasonal variation of circulating levels of sexual hormones than in most other cervids. In that case, either other hormones than those investigated in this study are involved.
or the final regulation of the antler cycle occurs at the tissue level.

We would like to thank Don Hartbauer and the staff at the Animal Quar ters and Bill Hauer and the staff at the Large Animal Research Station, University of Alaska, Fairbanks for their assistance in maintenance and sampling of the reindeer, Peter Bubenik and Yoko Imai for the preparation of data and the manuscript and Dr. D. Bolt (USDA Animal Hormone Program, Beltsville, MD, U.S.A.) for the generous gift of pituitary hormones. The project was supported by NSERC Canada, Institute of Arctic Biology and the Granting Agency of the Czech Republic (grant #505/95/029).

References


