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Brief communication

Progesterone during the breeding season and pregnancy in female muskoxen on different dietary regimens

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Abstract: Previously, we documented lowered calving success in captive muskoxen taised for 6 years on a low nutritional plane. In an effort to identify causes of lowered calf production, we looked at serum progesterone during the breeding season in muskoxen raised on either a high (HP) or low (LP) nutritional plane. Complete cessation of estrous cyclicity in 2 parous cows was the only irregularity identified. Abortion and/or embryonic loss occurred in both HP and LP cows. We also compared progesterone during pregnancy between the 2 nutritional planes and lactating and non-lactating cows. The timing of the rise and fall of the mid-pregnant progesterone peak was consistent with stage of gestation and apparently independent of time of breeding, diet or lactational status.

Key words: estrous cycle, body condition, embryonic loss, abortion.

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Introduction

There is now good evidence that habitat quality from mid-August to mid-October affects the ability of female muskoxen to regain sufficient body weight for successful reproduction (White *et al.*, 1989; Parker *et al.*, 1990; Adamczewski *et al.*, 1992; White *et al.*, 1997). Under good conditions muskoxen can calve annually and produce their first calf at 3 years of age (Latour, 1987; Rowell, 1991). Calving as a 2-year-old does occur, both in the wild and captivity, but is less common (Jingfors & Klein, 1982; Rowell *et al.*, 1987).

Under captive conditions, the probability of a cow calving falls below 50% when autumn body weight is less than 180 kg and body condition score (on a scale of 3-15) is less than 6 (White *et al.*, 1997). These authors recorded calving success in 2 groups of muskoxen maintained for 6 years on either a high (HP) or a low (LP) nutritional plane and were able to document a clear depression in calving success among LP cows. We, therefore, wanted to know if there were differences between the 2 groups in estrous cycle length or evidence of repeat cycles, establishment of pregnancy or indication of embryonic loss. In 1992, in conjunction with the study on calving success, we began collecting weekly blood samples during the breeding season from the most tractable animals within each nutritional group. In addition, plasma samples were collected throughout pregnancy (1993/94) in 3 HP and 3 LP cows to compare the pattern of progesterone secretion over mid to late pregnancy in both lactating and non-lactating females. This brief report is a summary of these results.

Methods

As part of a long term nutritional study in muskoxen, females ranging in age from 2.5-7.5 years-old at the time of breeding, have been maintained on either a high plane or a low plane of nutrition since 1987. High plane animals were allowed to graze pasture, had year-round access to brome-grass hay and were supplemented with a pelleted ration twice weekly (Quality Texture, Fisher Mills, Seattle; Alaska Mill and Feed, Anchorage) to give a daily pellet dry matter intake of 14 $g/kg^{0.75}$. Low plane cows were given slightly restricted grazing during the summer and allowed free access to brome-grass hay. They did not receive the pellet supplement and, in winter, had access to hay only. Body weight (BW) was measured weekly during the mating season (August 1-October 31) and body condition score (BCS) was assessed twice-monthly in August and September. Body condition score, a subjective index of subcutaneous fat and tissues, was determined from three regions on the animal: 1) the back (immediately caudal to the withers), 2) the ribs and 3) over the tuber coxae (pins) (Gerhart *et al.*, 1992; Gerhart *et al* , 1995). Each region was evaluated on a scale of 1-5, where 1=hide only and 5=maximum subcutaneous fat and other tissues (Gerhart *et al.*, 1995). Details on the nutritional regimen and condition scoring are given in White *et al.*, 1997.

An experienced breeding bull (> 4-years-old) was placed with each harem. The bulls were periodically exchanged between the HP and the LP group. Harems were set up in late Sept. in 1992, Aug. 17, 1993, and Aug. 22, 1994. All harems remained together until mid-late October. Throughout the breeding season weekly blood samples were collected by jugular venepuncture from a subset of HP and LP muskox cows (Table 1).

Table 1. Number of female muskoxen sampled weekly in each breeding season, at the Large Animal Research Station, Fairbanks AK. The females were divided between a high (HP) or low (LP) nutritional plane.

	1992	1993	1994	TOTAL
HP	1	3	4	8
LP	5	4	4	13

Jugular blood samples were collected from 3 HP and 3 LP pregnant muskoxen throughout the winter of 1994/95. Three of these cows (2HP, 1LP) were lactating and in 3 cows (1 HP, 2 LP) lactation had ceased before or during the breeding season. Plasma samples were collected at 1-2 week intervals after the breeding season. The plasma was harvested within 2 hours of collection and remained frozen until assayed.

Progesterone was determined by RIA. In 1992 samples were assayed by Reproductive Endocrine Labs, Colorado State University, Fort Collins, Colorado (Niswender, 1973). Assay sensitivity was 0.05 ng/ml. In 1993 and 1994 samples were assayed with commercial radioimmunoassay kits (Diagnostic Products Corporation; Los Angeles, CA). Sensitivity of the assays was 0.03 ng/ml. Intra-assay CV's were 4.9% (low) and 6.3% (high) in 1993 and 2.2% and 8.1% (low) and 9.5% and 16.2% (high) in 1994. All samples for each year were run in a single assay. The use of RIA to successfully assay steroids from muskox plasma has been demonstrated (Rowell & Flood, 1988; Rowell *et al.*, 1993). To validate the immunoassay kits, a subset of duplicate aliquots spanning the expected period of the progesterone rise and decline during pregnancy were submitted to Reproductive Endocrine Labs in Colorado for separate analysis. The results from the independent lab and the immunoassay kits corroborated very well.

The care and handling of the animals (and all study protocols) were approved by an independent animal welfare committee.

Results and discussion

Progesterone during the breeding season

Among the sampled females, all HP cows calved in 1993 and 1994 but only 3 of 4 calved in 1995. Two of 5 LP cows did not calve in 1993, one failed to calve in 1994 and again in 1995. Failure to ovulate (progesterone never rose above 0.5 ng/ml throughout the breeding season) was the mechanism employed by the 2 LP cows that failed to calve in 1993. One of these cows was not lactating while the other was suckling a yearling calf. The September BCS of both cows was 4 and we consider it probable that the cessation of estrous activity was associated with their very poor body condition. A similar situation was identified in field collections where the ovaries of 2 parous, female muskoxen had no corpora lutea or other indications of cyclic activity at the end of October (Rowell *et al.*, 1987). Reduced or absent courtship activity in the breeding season following a severe winter could also reflect the absence of estrous cycles (Gray, 1987).

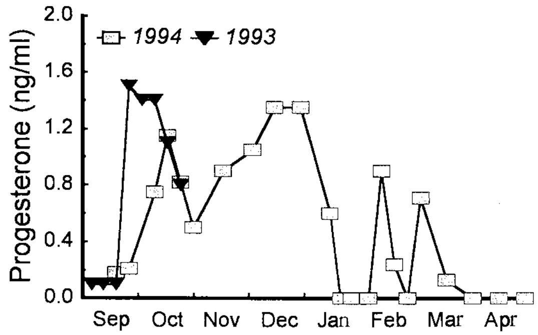

Fig. 1. Progesterone profiles for a female muskox that underwent abortion/embryonic loss in 2 consecutive breeding seasons. In 1993 sampling stopped shortly after harems were broken up. At this point the cow's (Sine) progesterone had remained elevated for 5 consecurive weeks but she failed to calve the following spring. In 1994 physical signs of an abortion were apparent at the end of December when progesterone returned to baseline. Following the abortion Sine resumed cyclic activity which continued into March.

It has been suggested that early embryonic loss/abortion may be a mechanism for controlling female productivity among muskoxen (Adamczewski, 1995) and in this study abortion and/or embryonic loss occurred on 3 occasions in 2 cows, 1 LP and 1 HP. In the LP cow (Sine), elevated progesterone levels (>0.5 ng/ml) were noted for 5 consecutive weeks in 1993 and for 10 consecutive weeks in 1994 (Fig. 1). In November, 1994, ultrasound corroborated the interpretation of ptegnancy. Physical evidence of abortion was only evident in 1994 although no fetus was recovered. This cow had not calved since 1991. Het fall BW was above 180 kg (207 kg-1993, and 225 kg-1994) but in both years her BCS was around the threshold value of 6 (5.6 - 1993 and 7.4 - 1994).

There are 2 equally plausible interpretations for the early embryonic loss, ecological or pathological:

Ecological - Sine's body condition in 2 consecutive autumns was marginal (BCS at threshold, BW above threshold). She was able to conceive but body condition did not improve and/or was not adequate for mid-winter conditions and somewhere between November/December she aborted or reabsorbed her fetus.

Pathological - This cow has a subclinical condition inhibiting adequate placental formation. Endometrial scarring has been observed in captive muskoxen (Blake, unpubl. obs.). This condition does not produce overt clinical symptoms but can seriously impair placental establishment. We know that the muskox corpus luteum of pregnancy regresses completely by December (Rowell *et al.*, 1993), and, if at this time the placenta is small and underdeveloped, the risk of abortion would be very high. The fact that this cow is 12 years old and has aborted in consecutive years also supports a pathological interpretation.

We cannot discriminate between the 2 explanations. This cow died of unrelated causes before the issue could be resolved.

The HP cow showing signs of early embryonic loss (Unni) had elevated progesterone for an estimated 6 weeks in 1994 and it is assumed she was pregnant. By early November progesterone had declined to baseline, ultrasonography indicated she was not pregnant and a large preovulatory follicle was visible on one ovary. Early pregnancy loss in Unni, a HP cow, is harder to explain in terms of a controlling mechanism for female productivity. Both BCS (12.3) and BW (239 kg) were increasing from August to October and remained above theoretical thresholds for successful breeding. This cow is 11 years old and, in her case, we could hypothesize an age effect or possibly a subclinical condition. She calved successfully the following year.

Early pregnancy loss occurs in all species and is associated with a complex interaction of physiological and genetic elements (Austin, 1972). These preliminary data can neither support nor refute the suggestion that early pregnancy loss is a mechanism controlling female productivity. However, it does emphasize some of the variables that should be considered when invoking controlling mechanisms and the need for a reasonably large sample size before conclusions can be drawn. It is interesting to note that following pregnancy termination both cows apparently resumed estrous cycles (Fig 1). Continuous cyclic activity into March has been observed before in well conditioned, non-pregnant, captive muskoxen (Rowell, 1991) and the occasional report of a very late born calf suggests that it must also happen in the wild (Alendal, 1971). However, this would not be an expected result if poor body condition had precipitated the abortion.

Progesterone during pregnancy

In all 6 muskoxen progesterone began to rise at 10-12 weeks gestation to levels 2-3 times those normally found during the estrous cycle. Progesterone then dropped suddenly between 19-22 weeks gestation, returning to levels typical of the estrous cycle

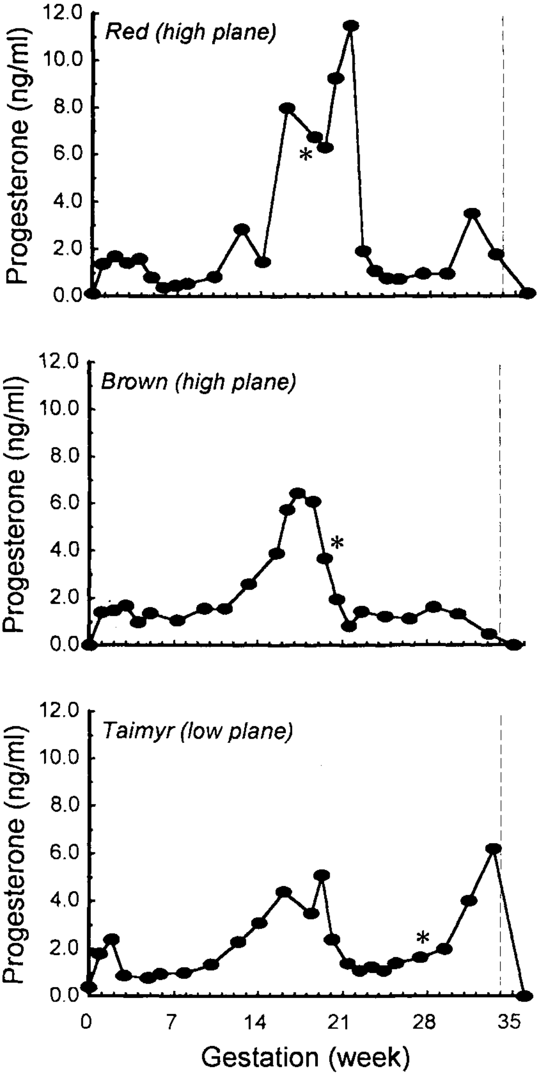

Fig. 2. Progesterone profiles from 3 individual muskoxen throughout pregnancy. All cows (2 HP and 1 LP) were lactating during rhe first half of gestation. Weaning is indicated by the asterisk and calving by the dotted line.

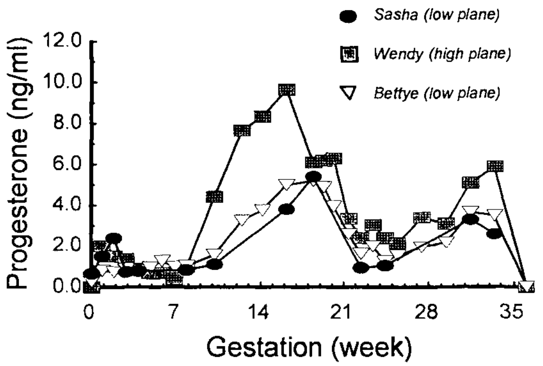

Fig. 3. Individual progesterone profiles throughout pregnancy from 3 non-lactating muskoxen (1 HP and 2 LP).

(Figs. 2 and 3). This pattern of progesterone secretion has been previously described (Rowell *et al.*, 1993) but its uniqueness among ruminants makes it important to demonstrate repeatability of the profile in diffetent muskoxen undet different conditions.

The mid-pregnant peak of progesterone occurred at the same stage of gestation (19-22 weeks) in both HP and LP cows regardless of whethet or not they were lactating (Figs. 2 and 3). This unusual profile was first documented in captive, non-lactating muskoxen on a high nuttitional plane Rowell *et al.*, 1993). Because natural weaning tends to occur at approximately the same time as the mid-pregnant progesterone peak, we were looking for a temporal association between the two events. The timing of weaning did not show any consistent relationship with the peak or decline of progesterone. The time of the progesterone peak in relation to the stage of gestation was consistent in both studies regardless of the date of breeding. This implies a physiological event related to the fetus and/or placenta.

We report here some of our observations on the characteristics of the estrous cycle in muskox cows that have been reared for over 6 years on either a high or low nutritional plane. We found a complete cessation of estrous cyclicity during the breeding season in 2 matute, parous cows in very poor body condition. No other irregularities of the estrous cycle were evident in our limited sample. We have documented abortion and early embryonic loss in both a HP and LP cow, although the role this may play in regulating productivity remains unclear.

Progesterone profiles during pregnancy support and expand previous documentation of a completely unique pattern. The timing of the progesterone rise and fall is remarkably consistent with stage of gestation and apparently independent of timing of breeding, diet or lactational status. This suggests an endocrine event that is a reflection of fetal/placental development rather than the maternal environment.

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