LEVELS OF SOME REPRODUCTIVE HORMONES IN RELATION TO PREGNANCY IN MOOSE: A PRELIMINARY REPORT

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Abstract: Blood samples were collected from two captive yearling female moose (Alces alces andersoni) from August 1983 to June 1984. The cows were bred by a yearling bull in November 1983. Blood sampling was conducted at weekly intervals. Serums were tested for levels of progesterone, leuteinizing hormone, and estrogen in relation to the pregnancy cycle. Progesterone levels were similar for both animals during estrus. One cow which produced twins had significantly higher progesterone levels throughout pregnancy compared to the cow with a singleton.

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Levels of reproductive hormones in blood serum has frequently been used to detect estrus cycles and pregnancy in cervids (Plotka et al. 1977), Verme and Ozoga 1981, Haigh et al. 1982). The determination of fetal production is often important for predictive models on population dynamics. A number of techniques including rectal palpation (Haigh et al. 1982), ultrasound (Smith and Lindzey 1982), and serum progesterone levels (Plotka et al. 1977) have been developed and used to this end. Rectal palpation and ultrasound both require experienced technical people and accuracy of results is influenced by the extent of fetal development. The estimation of pregnancy from progesterone levels in the serum is a laboratory procedure requiring the field investigator to collect blood from an adequate number of animals under proper conditions. The documentation of reproductive hormonal standards from anestrus to partuition in captive animals however greatly facilitates the interpretation of data obtained from wild captured animals.

McDonald (1980) described the hormonal control of the estrus cycle for a number of mammals. Two hormones from the anterior pituitary govern development of follicles in the ovary. Follicular stimulating hormone (FSH) and leuteinizing hormone (LH) are secreted continuously by the anterior pituitary throughout the cycle, but the

proportions and levels of each change during different stages. An ovulatory surge of LH is induced by rising estrogen concentrations which occur at estrus. Progesterone levels start to rise as the corpus luteum (CL) matures. If the CL is maintained due to pregnancy, progesterone levels remain high, but they decline toward the end of the cycle if no fertilization takes place. The current investigation provides preliminary insight into serum levels of LH, estrogen (E_2), and progesterone (P_4) for two captive moose from pre-estrus through to parturition.

METHODS

The study was conducted with 2 yearling moose, Niska and Daisy. Both animals were captured as week old calves and hand reared in a holding facility comprised of 4 seperate 1 ha enclosures. The moose had free access to a commercial pelleted ration and aspen browse throughout the year. The animals were conditioned to walk through a chute, and apart from being confined to the chute were not restrained or drugged. Blood samples (12 mls) were taken at 60 occasions between July 1983 and June 1984 from the cephalic veins of each moose, 33 prior to conception and 27 following. A yearling bull was introduced to the enclosure in mid October 1983. Precopulatory mounting was observed with Niska on November 5 and with Daisy

on November 17. Niska produced twin calves on June 7 1985 and Daisy had a singleton on June 20. Serum was harvested from the blood by centrifugation and stored at -20 C until assayed. Progesterone (P_4), estrogen (E_2), and leutenizing hormones were assayed by radio immunoassay. LH determination were for bovin LH.

Statistical analysis using a 2xn anova for paired comparisons were performed as described by Sokal and Rohlf (1969). Those blood samples (n=20) collected at similar stages during the pregnancy cycle (±2 days) were paired to test the null hypothesis that the levels of LH, E_2, and P_4 are similar for a cow supporting twin fetuses compared to a singleton.

RESULTS AND DISCUSSION

Levels of the three reproductive hormones are presented (Fig. 1). For both moose, fluctuations in the levels of LH and E_2 were evident prior to the onset of the first estrus cycle ie. 50-100 days prior to conception. The lack of any P_4 activity during this period discounts the possibility of any true estrus cycling. The first ovulation in pre-pubertal animals may in fact not be accompanied by estrus (Hansel and McEntee 1977).

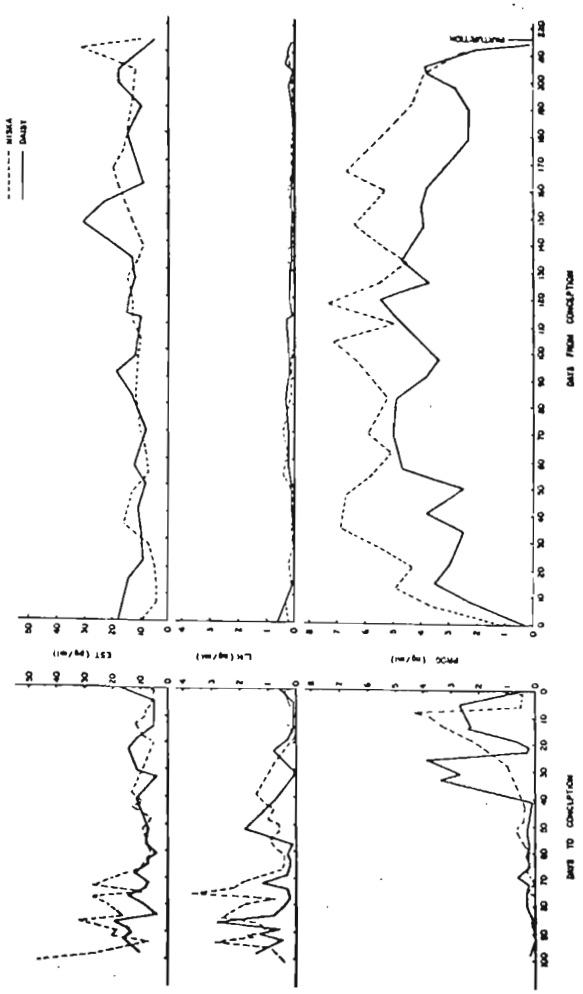

Figure 1. Progesterone (P_4), leutenizing hormone (L.H.), and estrogen (E_2) levels (prior to and during pregnancy) for 1 moose producing twin calves (Niska) and the other a singleton.

The 1st complete estrus cycle of Niska was initiated somewhere between October 9-17 when serum levels of P_4 exceeded 1.0 ng/ml; this increased to 4.3 ng/ml on October 29 and rapidly declined to 0.5 ng/ml by October 31. P_4 levels remained low through to November 5 when pre-copulatory mounting by the bull was observed. The estrus cycle was estimated between 18-26 days. Daisy experienced 2 estrus cycles between October 9 and November 17 each lasting 18-20 days. This compares to a range of 18-24 days for cattle, 14-19 days for sheep and goats (Hansel and McEntee 1977), and 21-30 days for white-tailed deer (Odocoileus virginianus) (Plotka et al 1977, Verme and Ozoga 1981). Sampling intensity was not sufficient to detect the anticipated peaks in LH during estrus. Levels of P rose sharply in both moose for the first 40-50 days following conception, after which they leveled out 40-50 days prepartum, at which time they started to decline. Niska had a higher mean P level of 5.24 ± 1.45 (1 S.D.) ng/ml throughout pregnancy than Daisy's 3.75 ± 0.96. Paired data analysis revealed the difference in P_4 between the 2 moose to be highly significant ($p \leq 0.001$), but there was also a large variance component ($p \leq 0.05$) among the individual moose which may represent genetic as well as environmental differences. Haigh et al. (1982) reported P_4 assays of 8.98 ± 2.2 ng/ml for 4 cows with twins compared to 6.82 ± 0.74 ng/ml for 11 cows with singletons in mid

March, approximately 160 days postpartum. Two of the cows judged to have twin fetuses had P_4 levels of 10.2 and 11.45 ng/ml with the remaining 2 at approximately 7 ng/ml. The range among cows with singletons was from 5.44 to 8.03 ng/ml. At this stage of pregnancy, P_4 levels for Niska were approximately 6.1 ng/ml and 4.0 ng/ml for Daisy. Plotka et al. (1983) reported lower plasma progestens for white-tailed deer which had been immobilized compared to deer which were manually restrained. They hypothesized that physical restraint induced stress resulting in adrenal progesterone contributions to the serum sample - thus the higher P_4 levels. They further suggested that anesthesia induced by drug administration had a relaxing effect on the adrenal by removing the source of stress; this may explain the elevated P_4 levels observed in the wild captured moose, for although they were anesthetized, capture was achieved by pursuit and darting from a helicopter. The stress associated with this form of capture has been known to cause capture myopathy in moose (Haigh et al. 1977). Plotka et al. (1983) reported that best results will be obtained only if the animal is completely drugged and sufficient time has elapsed to allow the adrenal secreted source to disappear from circulation. Another factor to consider is the decay of P_4 in whole blood; this occurs in cattle but not sheep (Rawlings, pers. comm.) There is an obvious need to standardize serum

harvest times and to test serum P_4 levels in drugged tame moose. Blood samples were not collected at consistent times following immobilization and the time of serum harvest from whole blood varied from 1 hour to 7 hours. These time spreads may have introduced substantial variation to the results obtained for the wild captured moose.

P_4 concentrations declined 40-50 days prepartum for both cows, dropping sharply 4-6 days prepartum. LH remained low throughout pregnancy ranging from 0 to 0.6 ng/ml. E_2 concentrations were 13.89 ± 9.46 pg/ml for Niska prior to conception and 12.00 ± 5.60 after compared to 10.62 ± 4.73 and 13.62 ± 5.46 respectively for Daisy. Statistical differences could not be detected.

SUMMARY

Our study has quantified levels of LH, E_2, and P_4 throughout the estrus cycle and pregnancy for moose. Progesterone levels for the cow carrying twin fetuses averaged 5.24 ± 1.45 ng/ml and 3.75 ± 0.96 in the cow with a singleton, and for paired comparisons a highly significant difference exists in P_4 levels between the 2 animals. The discrepancy in the levels reported for wild and tame animals raises some question about procedures associated with blood sampling from the wild. It is recommended that sufficient

time be allowed from time of immobilization to elapse for stress induced adrenal progesterones to disappear from the bloodstream. Harvesting of serum from whole blood should be carried out at standarized times.

Future efforts should concentrate on quantification of P_4 levels between females carrying twin and single fetuses, and, if so, when is the best time during pregnancy to sample. Continued studies will also contribute to our understanding of the hormonal control of the estrus cycle for moose, including the recurrence of the cycle in the absence of fertilization. Prolonged repetitive cycling may produce errors in the simple interpretation of P_4's to determine pregnancy.

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